Modified Nucleotides and RNA Origami in Nanotechnology and Synthetic Biology: A Survey

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Abstract

The interdisciplinary field of RNA nanotechnology, integrating principles from nanotechnology, synthetic biology, and chemistry, is advancing through innovations in modified nucleotides, RNA origami, and related technologies. Central to this progress is the development of RNA origami techniques, such as co-transcriptional folding, which enable the creation of functional RNA structures within biological systems, facilitating rapid evolutionary adaptation. Chemical modifications, especially in fully modified aptamers, enhance RNA stability and therapeutic efficacy, emphasizing the need for optimized pharmacokinetic properties. Theoretical advancements, including the SRAM method and scalable self-closing tubule structures, contribute to algorithmic self-assembly, offering dynamic control and enhanced yields through active self-disassembly. The non-reciprocal multifarious self-organization model further exemplifies innovative approaches to dynamic RNA systems, highlighting the potential for shape-shifting structures via programmable interactions. As the field evolves, addressing challenges in RNA origami and expanding its versatility remain crucial. The integration of RNA into in-situ origami structures for cellular applications promises to unlock new possibilities in medicine and biotechnology. Future research should focus on refining folding protocols, improving base-call accuracy, and expanding RNA technologies' applicability, aiming to harness the full potential of RNA nanotechnology across diverse scientific domains.

1 Introduction

1.1 Interdisciplinary Field Overview

RNA nanotechnology integrates principles from nanotechnology, synthetic biology, and chemistry, creating a robust platform for innovative applications. A key focus is RNA origami, which, despite its potential, remains underexplored in nanofabrication, highlighting the necessity for further research at the intersection of synthetic biology and nanotechnology [1]. This field promotes the development of synthetic strategies for the self-assembly of tubules, exemplifying the synergy between RNA nanotechnology and synthetic biology [2].

Structural design efforts aim to create 3D multicomponent self-assembling systems using allostery-mimetic building blocks, showcasing the interdisciplinary approach required for complex structure design [3]. Moreover, exploring non-reciprocal interactions in non-equilibrium dynamics seeks to enable automated control over self-assembled structures, drawing inspiration from the versatility of living systems [4].

Chemical modifications of nucleic acids, particularly through aptamer development, address critical limitations such as nuclease degradation and rapid renal clearance, which are essential for therapeutic applications [5]. Understanding RNA pseudoknot folding mechanisms is also crucial, as their thermodynamics and folding pathways significantly influence stability and biological functions [6].

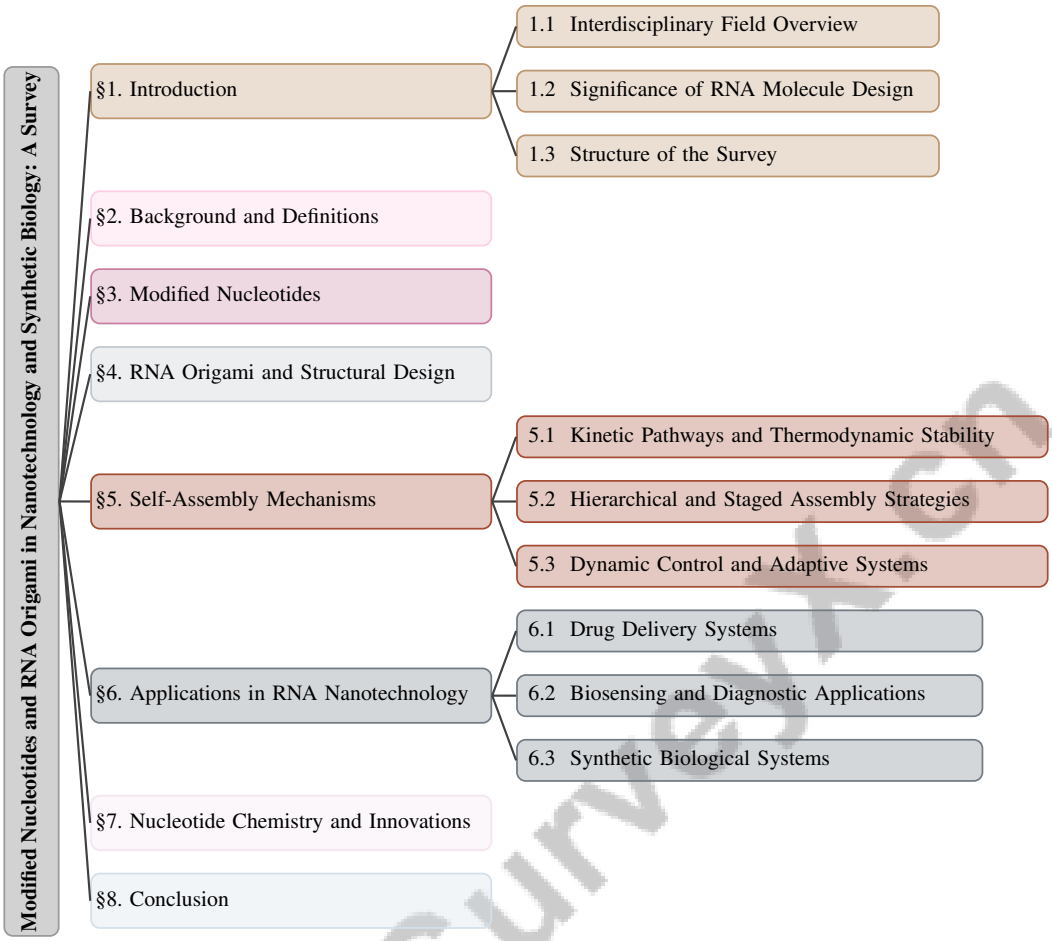

Figure 1: chapter structure

These elements collectively illustrate the rich interplay between nanotechnology, synthetic biology, and chemistry, advancing RNA-based systems' capabilities and applications. The interdisciplinary synergy between RNA and DNA nanotechnology enhances the design and functionality of nucleic acid-based systems, leading to innovative methodologies and applications. This collaboration facilitates the efficient construction of complex 2D and 3D nanostructures, such as molecular origami and hybrid RNA-DNA architectures, while expanding their potential in emerging biomedical applications, including targeted drug delivery and the stabilization of therapeutic RNA molecules. By leveraging unique properties of ribosomal RNA and messenger RNA as scaffolds, researchers can create stable and functional nanostructures, paving the way for advancements in biosensing, enzyme nanoreactors, and materials synthesis [7, 8, 9, 10].

1.2 Significance of RNA Molecule Design

The design of RNA molecules is critical for advancing therapeutics, nanotechnology, and synthetic biology, owing to RNA structures' inherent versatility and functional potential. Chemical modifications of RNA enhance stability and expand functional capabilities, effectively addressing challenges such as enzymatic degradation and limited chemical functionality [11]. For instance, nucleobase-modified antisense oligonucleotides for Duchenne muscular dystrophy demonstrate how chemical alterations can significantly improve exon-skipping efficiency [12].

In RNA nanotechnology, the stability and functionality of RNA nanostructures are paramount. Constructing stable RNA nanostructures is essential for applications like drug delivery and biosensing, where RNA's unique structural properties can create highly specific systems [8]. The use of long RNA scaffolds to fabricate complex 3D wireframe origami structures further underscores RNA's potential in biomedical applications [10]. Additionally, RNA design extends to developing complex

nanostructures based on programmable polymers, promising significant advancements in biotechnology [13]. Generating programmable forces in synthetic systems through RNA design offers flexibility and potential applications in nanotechnology [14].

Moreover, RNA's structural properties, such as stiffness, are critical in processes like encapsidation efficiency, where competition between linear and branched RNA forms influences assembly and stability [15]. Designing RNA molecules necessitates a comprehensive understanding of structural dynamics to optimize functionality across diverse applications. The demand for versatile RNA structures underscores the importance of RNA molecule design [1]. Proposed methods enhance selectivity for target structures in RNA assembly, highlighting the significance of RNA molecule design [2].

RNA modifications are also vital in gene expression regulation, necessitating novel detection chemistry to expand the list of detectable modifications [16]. The chemical and conformational diversity of modified nucleosides in tRNA affects structure, stability, and function, further emphasizing the role of chemical modifications in RNA design [17]. Additionally, leveraging RNA as a primary building block can simplify synthetic cell construction and enable evolutionary adaptation, illustrating RNA design's transformative potential in synthetic biology [18]. These advancements, coupled with precise engineering of molecular structures and diverse applications in nanotechnology, underscore the strengths of existing research in RNA molecule design [19].

1.3 Structure of the Survey

This survey is meticulously structured to explore the interdisciplinary field encompassing modified nucleotides, RNA origami, self-assembly, RNA nanotechnology, structural RNA design, and nucleotide chemistry. The introduction highlights the integration of nanotechnology, synthetic biology, and chemistry, emphasizing their collective significance in advancing RNA-based systems. Following this, Section 2 provides a comprehensive exploration of foundational concepts and critical definitions related to self-assembly pathways, underscoring essential terminologies that underpin molecular, nanoscale, and micron-scale self-assembly processes and their relevance to biological systems and medical applications [20, 11, 21].

Section 3 delves into modified nucleotides, elucidating their crucial roles in RNA design and functionality. It addresses diverse chemical properties and modifications that enhance RNA stability and functionality, drawing on recent findings that illustrate how these modifications contribute to structural diversity and dynamic behavior, particularly in transfer RNAs (tRNAs), which are extensively modified to optimize roles in protein synthesis and gene expression regulation. This section also explores the impact of modifications on RNA's electrochemical properties and interactions with macromolecules, underscoring the significance of modified nucleotides in cellular metabolism and therapeutic applications [5, 11, 22, 17, 16]. Section 4 scrutinizes RNA origami and structural design, discussing principles and techniques employed to create complex RNA structures with precise geometries, highlighting recent innovations in RNA-scaffolded origami.

Advancing to Section 5, the survey provides a comprehensive analysis of self-assembly mechanisms, delving into intricate kinetic pathways and thermodynamic stability that govern these processes, examining hierarchical and staged assembly strategies for constructing complex structures, and discussing systems designed for dynamic control and adaptation, including techniques for optimizing assembly protocols to circumvent kinetic traps and enhance yield [20, 23, 24, 25, 26]. Section 6 investigates diverse applications of RNA nanotechnology, including drug delivery systems, biosensing, diagnostic applications, and synthetic biological systems, illustrating the practical implications of theoretical principles discussed earlier.

In Section 7, the focus shifts to nucleotide chemistry and innovations, discussing recent advances in synthesis techniques, theoretical and computational approaches, and tools for RNA structure analysis. The survey concludes with Section 8, synthesizing the key points discussed and reflecting on the current state and future prospects of this vibrant interdisciplinary field. This structured approach facilitates a coherent presentation of information, effectively navigating the complex domain of RNA nanotechnology, which encompasses the innovative use of ribosomal RNA as a scaffold for constructing stable 2D and 3D origami nanostructures, while highlighting diverse applications in biomedical fields, including drug delivery and the development of artificial ribozymes [7, 10].The following sections are organized as shown in Figure 1.

2 Background and Definitions

2.1 Concept of Modified Nucleotides

Modified nucleotides are crucial in enhancing RNA design and functionality, offering a versatile toolkit for improving RNA molecules' structural and functional properties. These chemical modifications stabilize RNA structures and expand their therapeutic potential by addressing challenges such as nuclease degradation and rapid clearance, thereby improving the stability and efficacy of nucleic acid-based therapies [17, 5]. In tRNA, modified nucleosides are essential for protein synthesis, providing structural and functional enhancements [17]. In therapeutic applications, these modifications enhance aptamers and RNA therapeutics by improving binding affinity and specificity, leading to better therapeutic outcomes [5]. However, detecting endogenous modified nucleoside triphosphates in mammalian cells remains challenging due to their low abundance and poor ionization efficiency in mass spectrometry, necessitating sensitive detection methods [27].

In synthetic biology, modified nucleotides facilitate the creation of synthetic cells capable of self-replication and evolution, integrating genetic information with molecular machinery [18]. This potential is further exemplified in DNA origami technology, where modified nucleotides enhance the design of intricate RNA nanostructures [19]. By manipulating RNA's chemical properties, researchers can engineer RNA origami structures that self-assemble into predetermined shapes and functionalities [1]. The integration of modified nucleotides not only strengthens RNA's stability and functionality but also fosters innovative applications across various scientific domains.

2.2 RNA Origami and Structural Design Principles

RNA origami and structural RNA design leverage RNA's ability to fold into diverse shapes, enabling the construction of complex nanostructures with precise geometries and functionalities. These principles, grounded in RNA's modularity and programmability, allow for dynamic reconfiguration of components, enhancing adaptability for specific tasks [28]. Techniques such as using RNA scaffolds with DNA staples to form complex three-dimensional structures exemplify RNA origami's foundational principles, where RNA serves as the primary scaffold and DNA staples stabilize the structures [10, 7]. The oxRNA model provides accurate simulations of RNA dynamics, accounting for structural and electrostatic factors, particularly in viral contexts, emphasizing the importance of considering physical and chemical interactions in RNA origami design [29].

Categorizing DNA origami methodologies into design, assembly, and application stages offers a framework adaptable to RNA origami, facilitating systematic development of RNA-based nanostructures [19]. These advancements enhance the capability to create sophisticated RNA nanostructures, broadening RNA's potential applications in nanotechnology, therapeutics, and synthetic biology.

2.3 Self-Assembly in RNA Nanotechnology

Self-assembly in RNA nanotechnology involves the spontaneous organization of RNA molecules into ordered structures, driven by intrinsic molecular interactions. This process is essential for constructing complex nanostructures with applications in drug delivery, biosensing, and synthetic biology. Modeling the self-assembly of viral particles, especially those with multipartite genomes, reveals intricate dynamics [30]. RNA stiffness significantly affects encapsidation free energy and virus assembly efficiency, complicating the understanding of self-assembly mechanisms [15].

The Staged RNA Assembly Model (SRAM) proposes using RNAse enzymes to enhance assembly efficiency, offering a novel approach to overcoming self-assembly complexities [31]. However, existing methods often yield interactions that are overly complicated for practical implementation, highlighting a gap between theoretical designs and experimental capabilities [32]. The reliable assembly of complex 3D structures from a limited number of distinct building blocks remains challenging, often leading to misassembled states and reduced yields [3].

The economical self-closing assembly (ESCA) method addresses these challenges by employing multi-component systems to enhance selectivity and minimize off-target structures [2]. Additionally, active self-disassembly, inspired by biological systems, has been proposed to improve the yield of self-assembled structures, offering a dynamic approach to self-assembly [33]. Optimizing yield and

kinetics in self-assembly processes is essential for creating complex structures with thousands of components, necessitating a comprehensive understanding of the underlying pathways [34].

The folding mechanisms of RNA pseudoknots, crucial for various cellular functions, remain poorly understood regarding their thermodynamics and the influence of ionic conditions on assembly pathways [6]. Encoding desired structures through interactions while maintaining the ability to retrieve and transition between these structures presents a significant challenge in RNA self-assembly [4]. These insights emphasize the intricate balance of interactions required to achieve targeted structural formations, paving the way for advancements in RNA nanotechnology applications.

In recent years, the study of modified nucleotides has gained significant attention due to their critical role in RNA stability and functionality. Understanding the various types of modifications and their chemical properties is essential for advancing RNA research and applications in nanotechnology and therapeutics. Figure **??** illustrates the hierarchical classification of these modified nucleotides, focusing on their chemical properties, modification types, and the implications of these factors on RNA behavior. This figure not only highlights advanced assembly techniques and detection methods but also emphasizes the importance of precise engineering in the design of RNA for innovative applications. By integrating these insights, we can better appreciate the complexity and potential of modified nucleotides in contemporary scientific research.

Figure 2: This figure illustrates the hierarchical classification of modified nucleotides, focusing on chemical properties, modification types, and their impact on RNA stability and functionality. It highlights advanced assembly techniques, detection methods, and the role of precise engineering in RNA design for nanotechnology and therapeutic applications.

3 Modified Nucleotides

3.1 Chemical Properties and Modification Types

Exploring the chemical properties and modifications of nucleotides is crucial for enhancing RNA's stability and functionality, which are vital for nanotechnology and therapeutic applications. Nucleobase-modified antisense oligonucleotides improve duplex stability and RNA binding affinity, facilitating effective exon-skipping, a significant therapeutic advancement [12]. These modifications counteract RNA's instability and vulnerability to ribonuclease degradation, as shown by hybrid origami structures that exhibit greater resistance than traditional DNA–RNA duplexes [8]. Fully modified oligonucleotides, such as those with 100% 2-O-Methyl and 2'-fluoro modifications, are noted for their enhanced stability, essential for maintaining functional integrity in biological settings [35]. Co-transcriptional RNA origami innovations enable the direct production of RNA structures within synthetic cells, eliminating the need for pre-formed DNA structures and allowing dynamic cellular functions, thereby demonstrating the transformative potential of nucleotide modifications in synthetic biology [18].

Chemical modifications of nucleosides are categorized by their structural roles, as seen in tRNA, where specific modifications are essential for stability and function [17]. Innovative detection methods, such as 8-(diazomethyl) quinoline (8-DMQ) for labeling phosphate groups of NTPs, enhance sensitivity and facilitate the simultaneous detection of modified NTPs in mammalian cells [27]. Additionally, a chemistry-based enrichment approach in RNAseq library preparation enables the analysis of m7G and m3C modifications, expanding RNA modification detection capabilities [16]. Challenges like the limited availability and stability of RNA scaffolds for origami constructions highlight the need for stable and versatile designs [7]. Size limits, stability issues, and production scalability in DNA origami also present significant challenges relevant to RNA origami [19]. These advancements underscore the importance of precise engineering in RNA design, enabling the creation of RNA molecules with tailored properties for specific demands in nanotechnology, therapeutics, and synthetic biology, thus broadening the scope and efficacy of RNA-based applications.

3.2 Enhancement of RNA Stability and Functionality

Enhancing RNA stability and functionality through chemical modifications is essential for advancing RNA-based technologies in nanotechnology and therapeutics. These modifications establish a robust

framework for improving RNA's structural integrity and functional capabilities, crucial for forming intricate nanostructures and facilitating complex biological processes. Modifications significantly enhance the precision and complexity of self-assembled RNA structures, stabilizing configurations and promoting sophisticated nanostructure formation [36]. Recent advancements in RNA assembly techniques have introduced methods that enhance stability and functionality by enabling rapid assembly without extensive thermal cycling, thus preserving structural integrity [1]. The use of periodic coloring patterns to control assembly processes further mitigates thermal fluctuations, contributing to RNA stability [2]. Additionally, allostery-mimetic polycubes, which activate or deactivate binding sites based on prior interactions, enhance assembly guidance, ensuring stable and functional RNA formations [3].

Figure 3: Hierarchical categorization of key strategies for enhancing RNA stability and functionality, focusing on RNA modifications, assembly techniques, and detection methods. Each category encompasses specific methods and innovations that contribute to the advancement of RNA-based technologies in nanotechnology and therapeutics.

Optimizing non-equilibrium self-assembly protocols represents a significant advancement over traditional fixed-parameter approaches, allowing for adaptive control of assembly conditions. This adaptability increases yields of both equilibrium and metastable structures, enhancing RNA assembly functionality [26]. Furthermore, active self-disassembly has been proposed to boost the yield of target structures beyond what is achievable through self-assembly alone, providing a dynamic approach to maintaining RNA stability and functionality [33]. Directing the folding process through interaction sequence design allows for the assembly of diverse geometries from fewer building blocks, demonstrating the efficiency and versatility of RNA modifications in structural design [37]. The exceptional sensitivity and specificity of AlkAniline-Seq for detecting RNA modifications, such as m7G and m3C, serve as powerful tools for profiling these modifications and elucidating their roles in enhancing RNA stability and functionality [16].

Integrating these sophisticated assembly strategies and detection methods is crucial for advancing RNA nanotechnology, paving the way for innovative applications in medicine and biotechnology. These developments highlight the critical role of precise engineering in RNA design, facilitating the creation of RNA molecules with customized properties tailored to meet the specific requirements of various applications, including the construction of stable RNA-based nanostructures for biomedical purposes, the development of RNA origami cytoskeletons for synthetic cells, and the exploration of RNA modifications influencing gene expression and functionality [18, 17, 7, 16, 10].

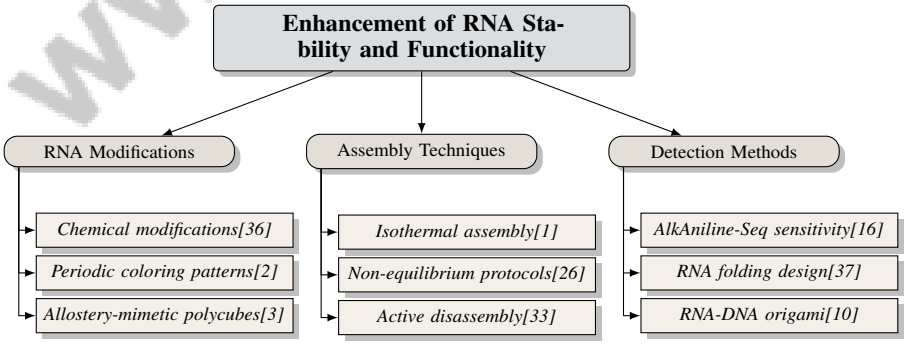

Figure 4: This figure illustrates the hierarchical categorization of key strategies for enhancing RNA stability and functionality, focusing on RNA modifications, assembly techniques, and detection methods. Each category encompasses specific methods and innovations that contribute to the advancement of RNA-based technologies in nanotechnology and therapeutics.

4 RNA Origami and Structural Design

4.1 Principles and Techniques of RNA Origami

RNA origami leverages RNA's modularity and programmability to construct complex nanostructures with precise geometries. This technique exploits RNA's unique folding properties to dynamically assemble intricate architectures. A significant advancement is the encapsulation of DNA templates and transcription machinery within giant unilamellar vesicles (GUVs), enabling co-transcriptional folding and assembly of RNA origami into functional structures [18]. Such methodologies demonstrate RNA origami's potential in synthetic biology, where controlled folding processes yield functional nanostructures.

The integration of DNA origami and RNA scaffolds is highlighted by the folding of ribosomal RNA into complex shapes using DNA oligonucleotide staples, enhancing the stability and functionality of the resulting structures [7]. This synergy underscores RNA origami's versatility in creating robust nanoscale architectures. Additionally, triangular DNA origami monomers with specified interaction patterns facilitate the construction of structures with precise geometrical features [2].

Initial assembly of RNA tiles into seed structures, which can be dynamically reconfigured using RNAse enzymes, exemplifies the adaptability of RNA origami for various applications [31]. Furthermore, DNA origami-based force clamps apply controlled mechanical forces to DNA, providing insights into transcription factor assembly dynamics and stability at the single-molecule level, which can be adapted for RNA origami techniques [38].

Modeling the interplay between RNA structure and capsid protein interactions using mean-field density functional theory emphasizes RNA stiffness's role in self-assembly, crucial for designing RNA origami structures that self-assemble with high precision [15]. Nanopore-based RNA reading techniques, which detect sequence and modifications based on deviations from canonical current patterns, offer novel insights into RNA folding dynamics and may verify the structural integrity of RNA origami constructs [22].

The principles and techniques of RNA origami significantly enhance the construction of intricate RNA nanostructures, utilizing abundant ribosomal RNA as scaffolds and integrating DNA oligonucleotides for stability and functionality. This progress broadens RNA applications in nanotechnology and synthetic biology, holding promise for innovative therapeutic strategies, including targeted drug delivery and RNA-based vaccines, thus laying the groundwork for potential use in emerging biomedical applications [7, 8, 18, 10].

As depicted in Figure 5, which illustrates the key techniques and applications of RNA origami, this innovative technique involves precise RNA folding into predetermined shapes, akin to paper origami, enabling the creation of complex nanostructures with diverse applications. The figure highlights various folding methods such as co-transcriptional folding, rRNA-DNA hybrid origami, and RNA-DX wireframe origami, alongside applications in synthetic cells, biomedical delivery, and RNA-based vaccines. Additionally, it addresses challenges like stability issues, RNA stiffness effects, and force-dependent interactions. Collectively, these examples underscore RNA origami's transformative potential in advancing synthetic biology and nanotechnology [8, 10, 18].

4.2 Innovations in RNA-Scaffolded Origami

Recent advancements in RNA-scaffolded origami techniques have expanded the potential for constructing complex and functional nanostructures. These innovations leverage RNA's unique properties to create scaffolds that facilitate precise nanoscale architecture assembly. A notable development is integrating DNA origami technology with single-molecule force spectroscopy, enabling measurement of force-dependent interactions in RNA structures [38]. This approach offers insights into RNA origami's mechanical properties, enhancing our understanding of RNA-based constructs' stability and dynamics.

Controlled mechanical forces applied to RNA scaffolds via DNA origami-based force clamps open new avenues for studying interactions within RNA assemblies, allowing researchers to investigate the effects of mechanical stress on RNA folding and stability [38]. The incorporation of force measurement capabilities into RNA-scaffolded origami represents a significant advancement in understanding RNA's mechanical behavior at the nanoscale.

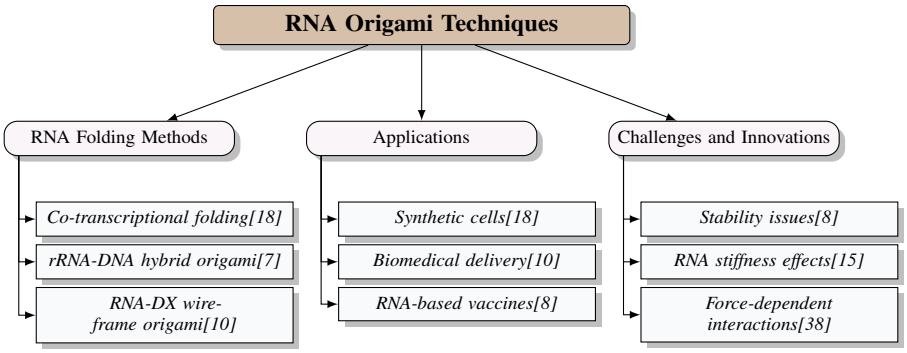

Figure 5: This figure illustrates the key techniques and applications of RNA origami, highlighting folding methods such as co-transcriptional folding, rRNA-DNA hybrid origami, and RNA-DX wireframe origami. The diagram also emphasizes applications in synthetic cells, biomedical delivery, and RNA-based vaccines, alongside challenges like stability issues, RNA stiffness effects, and force-dependent interactions.

Combining RNA scaffolds with DNA staple strands has led to developing hybrid nanostructures that utilize both nucleic acids' unique properties, enabling efficient construction of complex 2D and 3D origami shapes. This approach employs ribosomal RNA as a cost-effective and abundant scaffold, resulting in stable structures that exhibit enhanced resilience against ribonucleases and can be tailored for various biomedical applications, including targeted drug delivery and characterization of long RNA molecules [1, 7, 8, 10]. This hybrid approach facilitates constructing highly stable and functional nanostructures, essential for applications in drug delivery, biosensing, and synthetic biology. The precise control over RNA folding and assembly afforded by these techniques has the potential to revolutionize RNA nanotechnology, paving the way for innovative therapeutic and diagnostic tools.

Recent advancements in RNA-scaffolded origami techniques underscore RNA's transformative potential in nanotechnology. By leveraging RNA's modularity and programmability alongside DNA origami's structural precision, researchers can engineer advanced nanoscale devices for diverse applications, including targeted drug delivery, biosensing, and molecular machines, all while utilizing RNA scaffolds for stability and functionality in complex nanostructures. Recent studies highlight the potential of hybrid RNA-DNA origami in biomedical contexts, such as mRNA delivery and characterization of RNA structures, thus expanding the horizons of nucleic acid nanotechnology [19, 7, 8, 10]. These innovations enhance our understanding of RNA dynamics and expand the possibilities for engineering RNA-based systems with unprecedented functionality and complexity.

5 Self-Assembly Mechanisms

Exploring self-assembly mechanisms involves analyzing kinetic pathways and thermodynamic stability, which are crucial for the efficient and precise assembly of RNA nanostructures. The structural properties of RNA components, stabilizing agents like polyethylene glycol, and assembly conditions collectively influence the transition of disordered RNA into ordered nanostructures. RNA's stiffness, governed by base-pairing interactions, impacts encapsidation efficiency during viral capsid formation, while assembly pathways—nucleation versus aggregation—are pivotal in determining kinetics and outcomes. Understanding these dynamics is essential for optimizing RNA-based nanomaterials in biomedical applications [15, 7, 2, 39].

5.1 Kinetic Pathways and Thermodynamic Stability

Kinetic pathways and thermodynamic stability are key in governing the self-assembly of RNA nanostructures, affecting the efficiency and precision of the assembly process. Self-assembly involves forming ordered structures from disordered components, balancing kinetic accessibility with thermodynamic favorability [20]. Optimizing this interplay to achieve desired structures within practical timescales remains challenging [20].

Techniques like the T-jump method enhance understanding of RNA folding pathways by capturing dynamic influences and ionic conditions, offering quantitative insights into self-assembly processes [6]. Theoretical models, including generalized Monte Carlo schemes, simulate self-assembly dynamics, allowing for monitoring structural transitions and error evaluation [4]. These simulations highlight the need for strategies that predict and control self-assembly.

Despite advancements, current methods often struggle to adapt dynamically to environmental cues, limiting practical applications of DNA and RNA nanostructures [40]. Innovative approaches that integrate kinetic and thermodynamic considerations are needed to enhance the adaptability and functionality of self-assembled RNA systems. Recent studies emphasize the critical interplay between kinetic pathways and thermodynamic stability, highlighting molecular interactions and energy minimization as vital for understanding complex structure formation, such as viral capsids, from RNA and protein components [20, 41, 42, 21, 39]. By synthesizing theoretical, computational, and experimental strategies, researchers can optimize these factors, enhancing RNA nanotechnology applications in drug delivery, biosensing, and synthetic biology.

5.2 Hierarchical and Staged Assembly Strategies

Hierarchical and staged assembly strategies are crucial for constructing complex RNA structures, systematically overcoming kinetic barriers and improving assembly efficiency. These strategies involve assembling smaller subunits into larger organized structures, effectively circumventing kinetic traps [24]. Machine learning enhances hierarchical assembly strategies by optimizing interaction potentials, maximizing both thermodynamic yield and kinetic accessibility through algorithms that iteratively train Gaussian process regression models [43].

As illustrated in Figure 6, the figure emphasizes the role of machine learning in optimizing interaction potentials, the use of linker-mediated assembly for programming interactions, and the construction of RNA nanostructures for diverse applications. This approach allows for the self-assembly of quasicrystals under broader conditions than previous methods, expanding experimental applications [44]. Adaptive optimization protocols dynamically adjust assembly conditions, leading to improved yields and reduced assembly times compared to static protocols [26]. Such strategies are essential for optimizing assembly processes and ensuring efficient formation of complex RNA structures.

Linker-mediated assembly offers a flexible method for programming interactions in colloidal systems, enabling the design of complex structures with fewer unique sequences [45]. This method draws inspiration from the natural folding of biopolymers, such as RNA and proteins, which achieve complex structures from limited building blocks [37]. Research on specifically sequenced DNA dendrimers demonstrates their ability to form amorphous gel structures, revealing a strong correlation between the fraction of bonded strands and material dynamics [46].

Integrating theoretical, computational, and experimental strategies in hierarchical and staged assembly enhances understanding of self-assembly processes and enables precise control over resulting structures and properties. By incorporating geometric design principles, advanced machine learning techniques, and sophisticated algorithmic optimization, researchers can significantly improve RNA assembly efficiency and accuracy. This advancement facilitates robust construction of complex RNA nanostructures, such as 2D and 3D origami, broadening RNA's potential applications in biomedical delivery, enzyme nanoreactors, and dynamic biomolecular assembly [7, 47, 31, 10].

5.3 Dynamic Control and Adaptive Systems

Dynamic control and adaptive systems in RNA assembly are crucial for developing reconfigurable nanostructures responsive to environmental changes and functional demands. These systems exploit self-assembly principles, allowing RNA structures to dynamically reconfigure through controlled interactions and environmental cues. For instance, using colloidal droplet chains with programmable DNA interactions facilitates folding into specific geometries, known as foldamers, showcasing potential for programmable assembly [37]. This adaptability is vital for applications requiring responsive behavior, such as smart drug delivery systems and adaptive biosensors.

Emerging trends in RNA nanotechnology highlight the importance of exploring far-from-equilibrium phenomena and developing dynamic theories to better predict and control self-assembly processes [20]. Integrating real-time control mechanisms enhances the functionality and adaptability of DNA

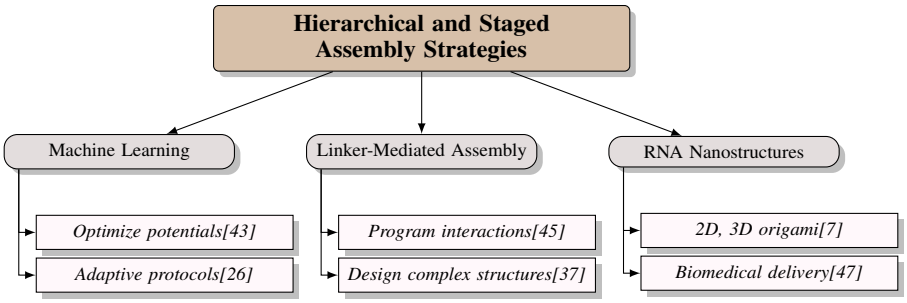

Figure 6: This figure illustrates the hierarchical and staged assembly strategies, emphasizing the role of machine learning in optimizing interaction potentials, the use of linker-mediated assembly for programming interactions, and the construction of RNA nanostructures for diverse applications.

nanostructures, providing a framework extendable to RNA systems [40]. This approach enables the construction of dynamic RNA assemblies capable of responding to external stimuli, opening new avenues for smart nanodevice design.

Kinetic-trap encoding offers an innovative method for encoding target structures during polymerization by manipulating the kinetics of component addition and removal. This technique leverages kinetic traps to enable rapid and precise retrieval of multiple target structures while addressing competition among partially formed fragments. By sculpting kinetic pathways rather than focusing solely on the free-energy landscape, this method enhances self-assembly efficiency and accuracy, facilitating the creation of complex structures from diverse building blocks, including synthetic DNA origami and protein complexes [48, 25]. This demonstrates the potential for dynamic systems to optimize both kinetic and thermodynamic objectives simultaneously, leading to improved self-assembly outcomes.

Additionally, multi-agent models employing the Lennard-Jones potential to simulate particle interactions in self-assembly, with temperature as a control input, further optimize assembly outcomes. This approach emphasizes the role of asymmetric interactions in controlling self-assembly, which could significantly impact bioengineering and disease mechanism understanding. The proposed dynamic bonding DNA model enhances DNA construct self-assembly simulations by integrating reversible hybridization and incorporating angular and dihedral interactions, capturing the collective effects of chemical structures on hybridization dynamics. This sophisticated framework facilitates detailed simulations of various self-assembly processes, including the kinetics of DNA tetrahedra and icosahedra formation and strand displacement operations essential for DNA computation [49, 47, 4].

Theoretical frameworks merging polymer physics and glass theory elucidate intricate folding mechanisms of proteins and RNA, establishing a comprehensive understanding of dynamic control processes in self-assembly. These frameworks reveal universal behaviors in biological systems, emphasizing the significance of quantitative descriptions in processes ranging from molecular folding to cellular communication, and demonstrating how programmable interactions can guide the formation of complex structures in synthetic materials inspired by natural biopolymers [37, 14, 21]. Future research should focus on refining models to incorporate realistic interactions and exploring their implications in diverse self-assembly contexts. Additionally, developing algorithms to simplify property predictions in flexible tile assembly model systems and investigating dynamic reconfigurability in self-assembling structures remain critical areas for advancement.

These efforts aim to deepen the understanding and practical implementation of dynamic control and adaptive systems in RNA nanotechnology, particularly through innovative uses of ribosomal RNA as scaffolds for molecular origami and programming dynamic assembly processes for viral proteins. This research lays the groundwork for novel applications in synthetic biology and nanomedicine by enabling the construction of stable RNA nanostructures and facilitating the development of RNA-based cytoskeletons within synthetic cells, ultimately paving the way for advanced solutions in biomedical engineering and therapeutic delivery systems [18, 7, 47].

6 Applications in RNA Nanotechnology

6.1 Drug Delivery Systems

RNA nanotechnology is crucial in advancing drug delivery systems by leveraging RNA's unique properties to construct precise and efficient delivery vehicles. Mini DNA–RNA hybrid origami nanobricks, for instance, demonstrate enhanced enzymatic resistance, making them robust carriers for therapeutic agents [8]. Their stability, augmented by chemical modifications, is vital for maintaining integrity in biological environments [35]. Frameworks like SAT-assembly facilitate the self-assembly of RNA-based nanostructures, optimizing kinetics and exploring particle variations to advance nanotechnology applications [50]. These frameworks enable precise engineering of tailored drug delivery systems.

Density functional theory (DFT) provides insights into the self-assembly of complex structures in binary mixtures, essential for controlling RNA-based delivery system properties [51]. Such understanding is critical for designing systems that effectively encapsulate and release therapeutic agents, enhancing efficacy. Moreover, active self-disassembly mechanisms improve assembly efficiency, offering dynamic approaches to optimize RNA nanostructure formation for drug delivery [33]. This dynamic control is crucial for developing responsive delivery systems adaptable to environmental cues.

The exploration of isotropic interactions in complex lattices, such as 2D Kagome and 3D diamond lattices, highlights RNA nanotechnology's potential in crafting intricate delivery vehicles [32]. These engineered structures optimize drug encapsulation and targeted release, enhancing therapeutic outcomes. Future research should focus on the effects of divalent ions on RNA folding pathways to refine experimental and computational design methods for RNA-based drug delivery systems [6]. By integrating these advancements, RNA nanotechnology can become a powerful tool for innovative drug delivery, enabling targeted and efficient therapeutic interventions.

6.2 Biosensing and Diagnostic Applications

RNA nanotechnology holds significant promise for biosensing and diagnostics due to its ability to form highly specific and stable structures. The modular nature of RNA allows for designing biosensors that detect a wide range of biological molecules with high specificity and sensitivity. RNA-based biosensors exploit unique folding properties to create platforms that undergo conformational changes in response to specific analytes, generating detectable signals [1].

A notable innovation involves RNA aptamers, which are engineered to selectively bind target molecules and serve as recognition elements in biosensors. Integrated into nanostructures, these aptamers enhance stability and binding efficiency, ideal for diagnostics [5]. Chemical modifications further improve RNA aptamers' resistance to degradation and functional capabilities, leading to robust diagnostic tools [17].

Integrating RNA nanotechnology with advanced detection methods like nanopore sequencing opens new avenues for biosensing applications. This approach enables real-time analysis of RNA modifications and sequences, crucial for diagnosing diseases and monitoring biological processes [22]. Computational models simulating RNA folding dynamics and interaction potentials optimize biosensing design, ensuring reliability and accuracy [4]. Hierarchical assembly strategies enhance RNA biosensors' complexity and functionality, enabling simultaneous detection of multiple targets [24].

As illustrated in Figure 7, the hierarchical structure of RNA nanotechnology applications in biosensing and diagnostics highlights key innovations such as RNA aptamers for target binding and stability, advanced detection methods like nanopore sequencing, and computational models. Additionally, it emphasizes hierarchical assembly strategies that contribute to enhanced complexity and multi-target detection.

RNA nanotechnology's diagnostic applications are exemplified by platforms for detecting viral pathogens, where RNA sensors' specificity and rapid response provide powerful tools for early diagnosis and infectious disease monitoring [6]. These advancements underscore RNA's transformative potential in biosensing and diagnostics, paving the way for innovative healthcare solutions. Continued exploration and refinement of RNA-based technologies can yield highly effective diagnostic tools addressing the growing demand for rapid and accurate detection of biological markers.

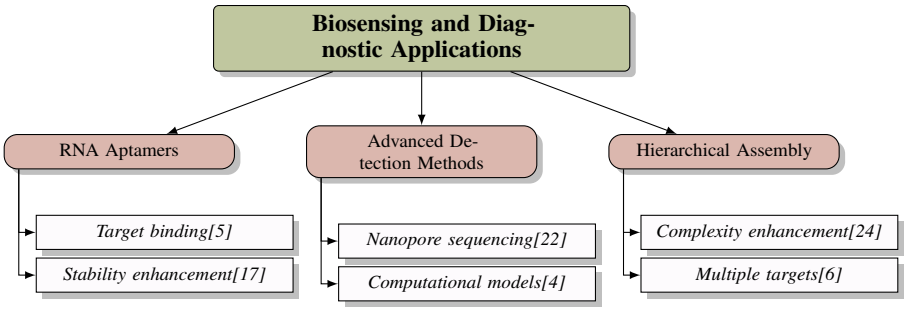

Figure 7: This figure illustrates the hierarchical structure of RNA nanotechnology applications in biosensing and diagnostics, highlighting key innovations such as RNA aptamers for target binding and stability, advanced detection methods like nanopore sequencing and computational models, and hierarchical assembly strategies for enhanced complexity and multi-target detection.

6.3 Synthetic Biological Systems

RNA nanotechnology is central to developing synthetic biological systems, offering innovative strategies for constructing complex and programmable architectures that mimic natural biological processes. Designing RNA-based structures with precise geometries and functionalities is essential for creating synthetic systems capable of performing specific biological tasks. The study by [2] illustrates RNA-based hardware's potential to simplify constructing synthetic life forms, facilitating evolutionary processes and expanding synthetic biology's scope.

The integration of RNA nanotechnology in therapeutic strategies is highlighted by using nucleobase-modified antisense oligonucleotides for exon-skipping, demonstrating RNA's versatility in developing targeted therapies within synthetic biological frameworks [19]. This approach underscores RNA nanotechnology's potential in addressing complex biological challenges through precise molecular design.

Linker-mediated phase behavior in DNA-coated colloids provides a flexible method for programming interactions in synthetic biological systems, enabling the design of complex structures with fewer unique sequences [45]. Inspired by natural biopolymer folding, such as RNA and proteins, this approach achieves complex structures from limited building blocks [37]. Specifically sequenced DNA dendrimers demonstrate the ability to assemble into amorphous gel structures, with a strong correlation between the fraction of bonded strands and material dynamics [46].

The SRAM method's application in developing synthetic biological systems allows for assembling infinite computable patterns, showcasing RNA nanotechnology's versatility in constructing intricate systems [31]. Additionally, findings that controlled nucleation and heterogeneous bond energies significantly improve self-assembly kinetics enhance RNA nanotechnology's capabilities in synthetic biology [3].

Future research should explore these findings' implications for other viral families and the effects of different RNA sequences on self-assembly, which could inform the design of more sophisticated synthetic biological systems. Developing more realistic models of RNA-capsid interactions, particularly focusing on excluded volume effects and RNA binding domains, will deepen understanding of RNA's role in synthetic biology [27].

These advancements highlight RNA nanotechnology's transformative potential in synthetic biology, providing a foundation for innovative systems with applications ranging from therapeutic interventions to user-defined materials with tailored properties. By harnessing RNA's distinctive properties, researchers are creating synthetic biological systems that closely replicate natural processes, enabling advanced solutions for pressing issues in medicine—where RNA origami can facilitate targeted drug delivery—and materials science, where RNA-based nanostructures enhance biomaterials' stability and functionality. This approach simplifies the design process by reducing the steps needed to translate genetic information into functional structures, opening new avenues for engineering complex molecular architectures with potential applications in therapeutics and nanotechnology [7, 18, 10].

7 Nucleotide Chemistry and Innovations

7.1 Innovative Synthesis Techniques

Method Name	Chemical Strategies	Structural Design	Functional Applications
IFRO[1]	Thermal Denaturation	Rna Origami Scaffold	Drug Delivery, Biosensing
ESCA[2]	Periodic Coloring Patterns	Triangular Dna Origami	Synthetic Biology
AAS[16]	Aniline Cleavage	Precise Mapping	Drug Delivery

Table 1: Overview of innovative synthesis techniques in RNA nanotechnology, detailing the chemical strategies, structural designs, and functional applications of various methods. The table highlights the IFRO, ESCA, and AAS methods, showcasing their unique contributions to drug delivery, biosensing, and synthetic biology.

Advancements in synthesis techniques are pivotal for RNA nanotechnology, enabling the creation of modified RNA with enhanced stability and functionality. Table 1 provides a comprehensive overview of the innovative synthesis techniques that are instrumental in advancing RNA nanotechnology, illustrating the interplay between chemical strategies, structural design, and functional applications. McKenzie et al. categorize non-native nucleic acid modifications by chemical structure and functional applications, aiding in the rational design of RNA molecules for specific uses [11]. The De Bruijn scaffold and staple strands represent significant progress in RNA origami, facilitating the construction of complex structures with precise geometries [1]. Multi-component systems and periodic coloring patterns further optimize RNA assembly, enhancing both efficiency and specificity [2].

Jacobs et al. offer a theoretical method for predicting nucleation barriers and assembly properties, providing insights applicable to various systems and enhancing understanding of RNA self-assembly [34]. The AlkAniline-Seq method, combining alkaline hydrolysis, dephosphorylation, and aniline cleavage, generates RNA fragments with unique 5'-phosphates, serving as a tool for analyzing RNA modifications [16]. Ni et al. categorize research on nucleic acid aptamers, focusing on modification strategies that influence stability, binding affinity, and therapeutic potential [5].

These innovative techniques underscore the importance of chemical strategies in RNA synthesis. By leveraging advancements in RNA origami and genetic encoding, researchers can engineer RNA molecules with enhanced structural stability and functional capabilities, leading to complex nanostructures for applications in targeted drug delivery, biosensing technologies, and synthetic biology [8, 18, 17, 7, 10].

7.2 Theoretical and Computational Advances

Theoretical and computational advances have significantly enhanced the understanding and design of RNA nanostructures, providing a framework for simulating RNA behavior. Recent developments in computational models and algorithms are crucial for investigating RNA folding dynamics, assembly, and interactions, especially for ribosomal RNA (rRNA), which constitutes a major portion of cellular RNA. These insights are pivotal for optimizing RNA-based systems, potentially leading to innovative biomedical applications [7, 21].

Computational algorithms simulate RNA folding pathways and interaction potentials, crucial for understanding thermodynamic and kinetic aspects of RNA self-assembly. These algorithms predict folding dynamics and identify stable configurations, facilitating the design of RNA nanostructures with enhanced stability and functionality. The integration of machine learning techniques into these models significantly improves prediction accuracy [7, 16, 31, 10].

Theoretical models, such as density functional theory (DFT) and molecular dynamics simulations, enhance the understanding of RNA's electronic and structural properties. Molecular dynamics studies reveal conformational dynamics of RNA fragments, showing how encapsidation alters structural motifs and interactions compared to freely-folding states. Electrostatic models elucidate interactions between positively charged protein tails and negatively charged RNA, stabilizing viral structures [52, 29, 7, 21, 15].

Computational methods enhance understanding of RNA-capsid interactions through simulations based on mean-field density functional theory, elucidating the encapsidation process. RNA stiffness, influenced by base-pairing and structural characteristics, affects the free energy associated with RNA

confinement within the capsid, providing insights into viral self-assembly and informing the design of virus-like particles [29, 9, 15].

Advanced computational tools, such as AlkAniline-Seq, improve profiling of RNA modifications like 7-methylguanosine (m7G) and 3-methylcytidine (m3C) at single nucleotide resolution, enhancing understanding of their roles in RNA stability, processing, transport, and translation [11, 16]. These tools provide a comprehensive view of RNA chemistry, facilitating identification of novel modification strategies that enhance RNA's therapeutic potential.

These advances deepen understanding of RNA dynamics and broaden potential applications in nanotechnology, therapeutics, and synthetic biology. By integrating computational design techniques with experimental methodologies, researchers can engineer RNA molecules as versatile scaffolds for constructing complex nanostructures, such as RNA origami, leveraging abundant rRNA and mRNA to create stable and functional nanostructures with significant potential for biomedical applications [7, 10].

7.3 Tools and Techniques for RNA Structure Analysis

RNA structure analysis is pivotal in RNA nanotechnology, providing insights into folding dynamics, stability, and functionality essential for constructing RNA-based nanostructures. Techniques like thermal annealing with complementary DNA oligonucleotides facilitate the creation of diverse polyhedral shapes with distinct properties. Advanced characterization methods, including dimethyl sulfate mutational profiling and cryo-electron microscopy, enhance understanding of RNA structures, crucial for biomedical applications such as drug delivery and artificial ribozymes [7, 10].

X-ray crystallography offers high-resolution images for precise determination of three-dimensional structures, complemented by nuclear magnetic resonance (NMR) spectroscopy, which provides insights into RNA's dynamic behavior in solution. These methods reveal conformational changes and interactions critical for understanding RNA's roles in cellular functions and applications [6, 29, 17, 7, 21].

Cryo-electron microscopy (cryo-EM) has revolutionized RNA structure analysis, particularly for large and complex assemblies, enabling visualization at near-atomic resolution without crystallization. This technique has facilitated exploration of RNA-based systems' intricate architecture and assembly mechanisms, leading to efficient construction of RNA origami using rRNA as scaffolds. Studies show these structures maintain integrity and stability under various conditions, expanding potential applications in biomedical fields through programmable assembly of viral proteins on DNA origami templates [7, 47, 9].

Alongside experimental techniques, computational tools are vital for RNA structure analysis, modeling RNA interactions, stability, and folding patterns critical for understanding ribosomal RNA and its applications. Molecular dynamics simulations and density functional theory (DFT) predict stable conformations and model RNA folding pathways, offering insights into thermodynamic and kinetic aspects of RNA assembly [7, 15].

High-throughput sequencing techniques, such as RNA-Seq, have transformed analysis of RNA modifications and interactions. Tools like AlkAniline-Seq enable detection and mapping of specific RNA modifications, providing detailed understanding of their impact on RNA stability and function [16]. These sequencing methods, combined with computational algorithms, facilitate comprehensive analyses of RNA structure and function, paving the way for innovative applications in therapeutics and synthetic biology.

Integration of experimental and computational tools in RNA structure analysis significantly advances understanding of RNA dynamics and interactions, as evidenced by studies on RNA pseudoknots and viral RNA encapsidation, revealing how factors such as RNA stiffness and ionic strength influence folding pathways and stability. This understanding facilitates design of sophisticated RNA-based systems, broadening applications across diverse scientific fields, including nanotechnology, biomedical delivery, and enzyme design [6, 29, 7, 15, 10].

8 Conclusion

RNA nanotechnology stands at the forefront of innovation, bridging the realms of nanotechnology, therapeutics, and synthetic biology. The integration of modified nucleotides and RNA origami has propelled the development of functional RNA structures capable of rapid adaptation, underscoring the transformative impact of RNA-based technologies on complex biological systems. Advances in co-transcriptional folding highlight the potential for creating sophisticated RNA architectures within living organisms, paving the way for novel therapeutic strategies.

The enhancement of RNA stability and therapeutic efficacy through chemical modifications, particularly in aptamers, remains a critical focus. Optimizing these modifications is essential to fully harness the therapeutic potential of RNA molecules. Expanding methodologies like AlkAniline-Seq will further illuminate the intricacies of RNA chemistry, broadening the scope of its applications.

Algorithmic self-assembly techniques, exemplified by the SRAM method, have achieved significant breakthroughs, enabling the construction of intricate structures with precision. The exploration of scalable, self-closing tubule architectures offers promising avenues for innovation in both nanotechnology and synthetic biology. Moreover, advancements in active self-disassembly techniques provide dynamic control over assembly processes, enhancing the yield and functionality of self-assembled structures.

Innovative models of non-reciprocal self-organization demonstrate the potential for dynamic, shape-shifting RNA systems, showcasing the versatility and adaptability of RNA constructs. Overcoming the challenges inherent in RNA origami and expanding the utility of these constructs are pivotal for future progress in the field.

The evolving landscape of RNA nanotechnology is characterized by its capacity to address contemporary challenges in medicine and biotechnology. By refining folding protocols and incorporating RNA into in-situ origami structures, researchers are poised to unlock new possibilities for RNA-based systems that emulate natural processes. As the field advances, improving accuracy in base-calling and expanding the applicability of RNA technologies will be crucial for realizing their full potential across diverse scientific domains.

References

[1] Emanuela Torelli, Jerzy Wieslaw Kozyra, Jing-Ying Gu, Ulrich Stimming, Luca Piantanida, Kislon Voïtchovsky, and Natalio Krasnogor. Isothermal folding of a light-up bio-orthogonal rna origami nanoribbon. *Scientific Reports*, 8(1):6989, 2018.

[2] Thomas E. Videbæk, Daichi Hayakawa, Gregory M. Grason, Michael F. Hagan, Seth Fraden, and W. Benjamin Rogers. Economical routes to size-specific assembly of self-closing structures, 2024.

[3] Joshua Evans and Petr Šulc. Designing 3d multicomponent self-assembling systems with signal-passing building blocks, 2023.

[4] Saeed Osat and Ramin Golestanian. Non-reciprocal multifarious self-organization, 2022.

[5] Shuaijian Ni, Houzong Yao, Lili Wang, Jun Lu, Feng Jiang, Aiping Lu, and Ge Zhang. Chemical modifications of nucleic acid aptamers for therapeutic purposes. *International journal of molecular sciences*, 18(8):1683, 2017.

[6] Jorjethe Roca, Naoto Hori, Yogambigai Velmurugu, Ranjani Narayanan, Prasanth Narayanan, D. Thirumalai, and Anjum Ansari. Monovalent ions modulate the flux through multiple folding pathways of an rna pseudoknot, 2017.

[7] Anastasia Shapiro, Noah Joseph, Nadav Mellul, Almogit Abu-Horowitz, Boaz Mizrahi, and Ido Bachelet. Folding molecular origami from ribosomal rna. *Journal of Nanobiotechnology*, 22(1):218, 2024.

[8] Lifeng Zhou, Arun Richard Chandrasekaran, Mengwen Yan, Vibhav A Valsangkar, Jeremy I Feldblyum, Jia Sheng, and Ken Halvorsen. A mini dna–rna hybrid origami nanobrick. *Nanoscale Advances*, 3(14):4048–4051, 2021.

[9] LaNell A. Williams, Andreas Neophytou, Rees F. Garmann, Dwaipayan Chakrabarti, and Vinothan N. Manoharan. Effect of coat-protein concentration on the self-assembly of bacteriophage ms2 capsids around rna, 2024.

[10] Molly F Parsons, Matthew F Allan, Shanshan Li, Tyson R Shepherd, Sakul Ratanalert, Kaiming Zhang, Krista M Pullen, Wah Chiu, Silvi Rouskin, and Mark Bathe. 3d rna-scaffolded wireframe origami. *Nature Communications*, 14(1):382, 2023.

[11] Luke K McKenzie, Roberto El-Khoury, James D Thorpe, Masad J Damha, and Marcel Hollenstein. Recent progress in non-native nucleic acid modifications. *Chemical Society Reviews*, 50(8):5126–5164, 2021.

[12] Bao T Le, Mick Hornum, Pawan K Sharma, Poul Nielsen, and Rakesh N Veedu. Nucleobase-modified antisense oligonucleotides containing 5-(phenyltriazol)-2-deoxyuridine nucleotides induce exon-skipping in vitro. *Rsc Advances*, 7(86):54542–54545, 2017.

[13] Jernej Rus. Antiparallel d-stable traces and a stronger version of ore problem, 2016.

[14] Dino Osmanovic and Elisa Franco. Generating forces in confinement via polymerization, 2024.

[15] Siyu Li, Gonca Erdemci-Tandogan, Paul van der Schoot, and Roya Zandi. The effect of rna stiffness on the self-assembly of virus particles, 2018.

[16] Virginie Marchand, Lilia Ayadi, Felix GM Ernst, Jasmin Hertler, Valérie Bourguignon-Igel, Adeline Galvanin, Annika Kotter, Mark Helm, Denis LJ Lafontaine, and Yuri Motorin. Alkanilineseq: profiling of m7g and m3c rna modifications at single nucleotide resolution. *Angewandte Chemie International Edition*, 57(51):16785–16790, 2018.

[17] Ville YP Väre, Emily R Eruysal, Amithi Narendran, Kathryn L Sarachan, and Paul F Agris. Chemical and conformational diversity of modified nucleosides affects trna structure and function. *Biomolecules*, 7(1):29, 2017.

[18] Mai P Tran, Taniya Chakraborty, Erik Poppleton, Luca Monari, Franziska Giessler, and Kerstin Göpfrich. Genetic encoding and expression of rna origami cytoskeletons in synthetic cells. *bioRxiv*, pages 2024–06, 2024.

[19] Swarup Dey, Chunhai Fan, Kurt V Gothelf, Jiang Li, Chenxiang Lin, Longfei Liu, Na Liu, Minke AD Nijenhuis, Barbara Saccà, Friedrich C Simmel, et al. Dna origami. *Nature Reviews Methods Primers*, 1(1):13, 2021.

[20] Stephen Whitelam and Robert L. Jack. The statistical mechanics of dynamic pathways to self-assembly, 2014.

[21] D. Thirumalai. Universal relations in the self-assembly of proteins and rna, 2014.

[22] Andrew M Smith, Miten Jain, Logan Mulroney, Daniel R Garalde, and Mark Akeson. Reading canonical and modified nucleobases in 16s ribosomal rna using nanopore native rna sequencing. *PloS one*, 14(5):e0216709, 2019.

[23] Jerome Durand-Lose, Jacob Hendricks, Matthew J. Patitz, Ian Perkins, and Michael Sharp. Self-assembly of 3-d structures using 2-d folding tiles, 2018.

[24] Stephen Whitelam. Hierarchical assembly may be a way to make large information-rich structures, 2015.

[25] Jim Madge, David Bourne, and Mark A. Miller. Controlling fragment competition on pathways to addressable self-assembly, 2019.

[26] Anthony Trubiano and Michael F. Hagan. Optimization of non-equilibrium self-assembly protocols using markov state models, 2022.

[27] Han-Peng Jiang, Jun Xiong, Fei-Long Liu, Cheng-Jie Ma, Xing-Lin Tang, Bi-Feng Yuan, and Yu-Qi Feng. Modified nucleoside triphosphates exist in mammals. *Chemical Science*, 9(17):4160–4167, 2018.

[28] S. E. Ahnert, I. G. Johnston, T. M. A. Fink, J. P. K. Doye, and A. A. Louis. Self-assembly, modularity and physical complexity, 2010.

[29] Giovanni Mattiotti, Manuel Micheloni, Lorenzo Petrolli, Luca Tubiana, Samuela Pasquali, and Raffaello Potestio. Molecular dynamics characterization of the free and encapsidated rna2 of ccmv with the oxrna model, 2024.

[30] Martin Castelnovo, Delphine Muriaux, and Cendrine Faivre-Moskalenko. Entropic control of particle sizes during viral self-assembly, 2013.

[31] Erik D. Demaine, Matthew J. Patitz, Robert T. Schweller, and Scott M. Summers. Self-assembly of arbitrary shapes using rnase enzymes: Meeting the kolmogorov bound with small scale factor (extended abstract), 2010.

[32] Erik Edlund, Oskar Lindgren, and Martin Nilsson Jacobi. Using the uncertainty principle to design simple interactions for targeted self-assembly, 2012.

[33] Karsten Kruse, Jean-Pierre Eckmann, and Wilson C. K. Poon. Active self-disassembly enhances the yield of self-assembled structures, 2024.

[34] William M. Jacobs, Aleks Reinhardt, and Daan Frenkel. Rational design of self-assembly pathways for complex multicomponent structures, 2015.

[35] Christina Kratschmer and Matthew Levy. Effect of chemical modifications on aptamer stability in serum. *Nucleic acid therapeutics*, 27(6):335–344, 2017.

[36] Daichi Hayakawa, Thomas E. Videbæk, Douglas M. Hall, Huang Fang, Christian Sigl, Elija Feigl, Hendrik Dietz, Seth Fraden, Michael F. Hagan, Gregory M. Grason, and W. Benjamin Rogers. Geometrically programmed self-limited assembly of tubules using dna origami colloids, 2022.

[37] Angus McMullen, Maitane Muñoz Basagoiti, Zorana Zeravcic, and Jasna Brujic. Self-assembly through programmable folding, 2022.

[38] Kevin Kramm, Tim Schröder, Jerome Gouge, Andrés Manuel Vera, Kapil Gupta, Florian B Heiss, Tim Liedl, Christoph Engel, Imre Berger, Alessandro Vannini, et al. Dna origami-based single-molecule force spectroscopy elucidates rna polymerase iii pre-initiation complex stability. *Nature communications*, 11(1):2828, 2020.

[39] Rees F. Garmann, Aaron M. Goldfain, and Vinothan N. Manoharan. Measurements of the self-assembly kinetics of individual viral capsids around their rna genome, 2018.

[40] Leopold N Green, Hari KK Subramanian, Vahid Mardanlou, Jongmin Kim, Rizal F Hariadi, and Elisa Franco. Autonomous dynamic control of dna nanostructure self-assembly. *Nature chemistry*, 11(6):510–520, 2019.

[41] Inbal Mizrahi, Robijn Bruinsma, and Joseph Rudnick. The spanning tree model and the assembly kinetics of rna viruses, 2022.

[42] Tao Hu and B. I. Shklovskii. Kinetics of viral self-assembly: the role of ss rna antenna, 2007.

[43] Sambarta Chatterjee and William M. Jacobs. Multi-objective optimization for targeted self-assembly among competing polymorphs, 2025.

[44] Aleks Reinhardt, John S. Schreck, Flavio Romano, and Jonathan P. K. Doye. Self-assembly of two-dimensional binary quasicrystals: A possible route to a dna quasicrystal, 2016.

[45] Janna Lowensohn, Bernardo Oyarzún, Guillermo Narvaez Paliza, Bortolo M. Mognetti, and W. Benjamin Rogers. Linker-mediated phase behavior of dna-coated colloids, 2019.

[46] Francis W. Starr and Francesco Sciortino. Model for assembly and gelation of four-armed dna dendrimers, 2005.

[47] Kun Zhou, Yihao Zhou, Victor Pan, Qiangbin Wang, and Yonggang Ke. Programming dynamic assembly of viral proteins with dna origami. *Journal of the American Chemical Society*, 142(13):5929–5932, 2020.

[48] Félix Benoist and Pablo Sartori. High-speed combinatorial polymerization through kinetic-trap encoding, 2024.

[49] Carsten Svaneborg, Harold Fellermann, and Steen Rasmussen. Dna self-assembly and computation studied with a coarse-grained dynamic bonded model, 2012.

[50] Lorenzo Rovigatti, John Russo, Flavio Romano, Michael Matthies, Lukáš Kroc, and Petr Šulc. A simple solution to the problem of self-assembling cubic diamond crystals, 2022.

[51] Alberto Scacchi, Maria Sammalkorpi, and Tapio Ala-Nissila. Self-assembly of binary solutions to complex structures, 2021.

[52] Tao Hu, Rui Zhang, and B. I. Shklovskii. Electrostatic theory of viral self-assembly: a toy model, 2007.

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