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Advancements in Aptamer-Based Nanoparticle Bioconjugates for Targeted Cancer Therapy: A Comprehensive Review

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ABSTRACT

Nanoparticle-aptamer bioconjugates are revolutionizing targeted cancer therapy. This review explores their transformative potential, tracing the evolution from monoclonal antibodies to versatile nucleic acid aptamers. We examine aptamer properties, functionalities, and the pivotal role of in vitro selection (SELEX). The integration of aptamers with drug-encapsulated nanoparticles is highlighted as a potent vehicle for targeted delivery. We scrutinize nanoparticle attributes, conjugation techniques, and clinical relevance while addressing challenges such as nuclease degradation and renal filtration. This comprehensive exploration underscores the promise of nanoparticle-aptamer bioconjugates in precision oncology, offering enhanced targeting and improved cancer treatment outcomes. Future directions, including combination with immunotherapy and overcoming drug resistance, are discussed.

Keywords: Aptamers, DNA, RNA, SELEX, Targeted Delivery

Highlights:

- Aptamers are short nucleic acid molecules that offer a promising alternative to monoclonal antibodies for targeted cancer therapy.
- Aptamers have several advantages over antibodies, including as being smaller in size and having improved tissue penetration, and reduced immunogenicity.
- Aptamer-drug conjugates (ADCs) allow for targeted delivery of therapeutic medicines to cancer cells, reducing harm to healthy tissues.
- Nanoparticles serve as diverse platforms for delivering aptamer-based therapies, and their characteristics can be manipulated to optimize drug delivery and improve the effectiveness of treatment.

- The SELEX technique is an effective method for extracting aptamers that have a strong attraction and specificity for specific compounds. This approach greatly aids in the creation of tailored treatments.

Introduction

The field of targeted cancer therapy has undergone significant advancements since the late 1990s, with the introduction of monoclonal antibodies such as Rituximab (Rituxan™), which received FDA approval in 1997. This breakthrough paved the way for the development and approval of numerous other antibody-based therapies such as Trastuzumab (Herceptin™, 1998), Bevacizumab (Avastin™, 2004), Cetuximab (Erbitux™, 2004), Mogamulizumab (Poteligeo™, 2012), Pembrolizumab (Keytruda™, 2014), Nivolumab (Opdivo™, 2015), Atezolizumab (Tecentriq™, 2016), and Tisagenlecleucel (Kymriah™, 2017), each targeting specific proteins or receptors on cancer cells [1-5]. However, these successes were tempered by the limitations inherent to antibodies, such as their large size, potential immunogenicity, and complex production processes [6]. Moreover, the complexities of biological development, including target antigen tolerance and batch-to-batch variability, pose significant challenges in the production of monoclonal antibodies [7].

In response to these challenges, researchers turned to nucleic acid aptamers, which are smaller, non-immunogenic molecules with high target specificity. Pioneering work by Szostak and Gold in in vitro evolution methodologies propelled aptamer development [8, 9]. Aptamers, which are short, single-stranded DNA or RNA molecules, possess benefits such as reduced size, lack of immunogenicity, and exceptional target specificity [10]. Aptamers have been utilized in controlled drug delivery systems, showcasing effectiveness in combating prostate cancer [11-13].

The limits of conventional cancer treatments, such as chemotherapy and radiation, which lack specificity and result in widespread side effects, emphasize the necessity for focused techniques [14]. Aptamers, with their ability to penetrate tumors, bind diverse targets, and exhibit minimal immunogenicity, offer a promising solution [15-19].

Aptamers have shown potential in theranostics, combining diagnostic and therapeutic functions [20, 21]. Modifications like locked nucleic acids (LNAs) enhance their stability and binding affinity [22, 23]. Aptamers can act as agonists, antagonists, or drug carriers, demonstrating versatility in therapeutic applications [24]. The functional characteristics and stability of aptamers are crucial for clinical applications. Strategies to improve stability include chemical modifications and incorporation into nanoparticles [25-27]. Aptamer-drug conjugates (ADCs) offer targeted delivery, enhanced efficacy, and design versatility [28-30]. The strategic design of aptamer-drug conjugates (ADCs) offers the potential to enhance targeted cancer therapy by improving drug delivery to specific cancer cells, thereby increasing therapeutic efficacy while minimizing off-target effects [31-33]

The integration of aptamers and nanoparticles for precise medication delivery is a notable breakthrough in the field of cancer treatment [34]. The integration of aptamers with drug-encapsulated nanoparticles is examined, highlighting their emergence as potent vehicles for targeted delivery. We scrutinize nanoparticle attributes, conjugation techniques, and clinical relevance while addressing challenges and future research directions. This comprehensive exploration underscores the promise of nanoparticle-aptamer bioconjugates

in ushering in a new era of precision oncology, offering enhanced targeting, controlled drug release, and improved cancer treatment outcomes [35].

In conclusion, aptamers hold immense promise for revolutionizing targeted cancer therapy. Continued research in this field is expected to yield innovative solutions for more effective and selective cancer treatments.

This review will explore the evolution of targeted cancer therapies, from monoclonal antibodies to aptamer-based approaches. We will examine the unique properties of aptamers, their selection process, and their integration with nanoparticles for drug delivery. The review will also examine present clinical implementations, obstacles, and prospective pathways in this swiftly advancing domain.

Limitations of Traditional Cancer Therapies

Although chemotherapy and radiation have long been fundamental in cancer treatment, they are frequently criticized for their non-specificity, resulting in widespread side effects that can greatly impact a patient's quality of life. These therapies do not differentiate between healthy and cancerous cells, resulting in damage to normal tissues and causing various side effects. This lack of specificity underscores the urgent need for more targeted therapeutic approaches that can distinguish between healthy and malignant cells, thereby minimizing collateral damage and improving patient outcomes [14].

Transition to Aptamer-Based Therapies

Monoclonal antibodies have had a profound impact on cancer treatment by providing a high level of precision in targeting antigens on cancer cells. Nevertheless, they possess inherent restrictions, including their substantial molecular dimensions that hinder tumor infiltration, and their capacity to provoke an immunogenic reaction, resulting in diminished effectiveness and possibly unfavorable responses. Moreover, the process of manufacturing monoclonal antibodies is intricate and expensive, which can restrict their accessibility and availability [36, 37].

In contrast, aptamers, which are small nucleic acid molecules, offer several advantages. Due to their reduced dimensions, they exhibit enhanced tumor infiltration and dispersion within the tissue. They have a lower probability of being identified as foreign by the immune system, thereby decreasing the likelihood of immunogenicity. Furthermore, aptamers can be chemically produced, providing benefits in terms of cost-effectiveness, scalability, and consistent quality across several batches. Aptamers has these characteristics, which make them a highly promising alternative in the field of targeted cancer treatments. They serve as a basis for the development of more efficient and less harmful therapeutic choices [38].

Aptamers: A Versatile Tool for Targeted Therapy

The exploration the unique advantages of aptamers as revolutionary tools for targeted therapy in cancer treatment.

Advantages of Aptamers over Traditional Antibodies

Aptamers offer several key advantages over traditional antibodies:

- **Smaller size and flexibility:** Aptamers, significantly smaller (20-60 nucleotides) than antibodies, can penetrate tumors more effectively, reaching target sites and enhancing therapeutic efficacy [15]. Their structural flexibility allows versatile binding across a broad spectrum of targets, crucial for effective therapeutic interventions [16].

Aptamers have a clear advantage over conventional antibodies since they are smaller in size, which improves their capacity to infiltrate tumors more efficiently. This attribute allows aptamers to reach and bind to target molecules within cancer cells more efficiently than larger antibody molecules, which may be impeded by the dense extracellular matrix of tumors. Zhu et al. (2022) conducted a study that showed aptamers to be more efficient than antibodies in penetrating solid tumors, resulting in enhanced targeting and therapeutic results [39].

Additionally, the reduced immunogenicity of aptamers, as highlighted by Zhou et al. (2017), minimizes the risk of adverse immune responses, making them safer for repeated administration in therapeutic contexts [40]. All of these features highlight how aptamers could be better cancer therapy targeting agents.

- **Enhanced tissue permeability:** Aptamers are able to permeate tissues more effectively than antibodies due to their tiny size, reaching tumor regions that are typically inaccessible to larger molecules [18].
- **Reduced immunogenicity:** Aptamers, resembling endogenous nucleic acids, exhibit minimal immunogenic responses, reducing the likelihood of adverse immune reactions—a significant advantage over protein-based therapeutic agents [19].
- **Cost-effective synthesis and minimal batch variability:** A key component of large-scale pharmaceutical manufacturing is the ability to produce aptamers with a low batch-to-batch fluctuation; chemical synthesis of aptamers enables this [17].

Aptamers for Theranostics

With its ability to combine diagnostic and therapeutic functions, aptamers have demonstrated great potential in the field of theranostics. Cancer therapy procedures might be made much more precise and effective with this dual functionality [21]. The theranostics potential of aptamers, which combine diagnostic and therapeutic capabilities in a single molecule, is becoming more and more apparent. A recent study investigated the potential of aptamer-based theranostics for targeted cancer treatment and real-time monitoring, offering hope for personalized medicine [20]. These theranostics agents can simultaneously detect disease biomarkers and deliver targeted therapy, offering a dual-function that enhances the efficiency of cancer treatment protocols.

Locked Nucleic Acids (LNAs): Enhanced Stability

Locked nucleic acids (LNAs) and other chemical changes have greatly improved the stability and binding strength of aptamers, hence increasing their efficacy in therapeutic applications. The alterations enhance the pharmacokinetic and pharmacodynamic characteristics of aptamers, allowing their application in many therapeutic contexts [22]. They have significantly improved the stability of aptamers in biological applications. Adding LNA to aptamer structures has been proven to greatly increase their ability to bind and withstand destruction by nucleases [23]. This enhancement not only improves the therapeutic efficacy of aptamers but also extends their circulation time in the bloodstream, offering prolonged therapeutic action.

Diverse Therapeutic Applications

Aptamers exhibit remarkable versatility in therapeutic applications, capable of acting as agonists, antagonists, or drug carriers. A study by Szymanowski et al. (2023) highlighted the multifunctionality of aptamers, demonstrating their capability to precisely target and modulate cancer cell functions [24]. Additionally, aptamers can be engineered to carry various therapeutic agents, showcasing their potential as versatile tools in targeted cancer therapy.

Aptamers possess a wide range of applications in cancer therapy owing to their capacity to selectively bind to a diverse array of targets:

Agonists or antagonists: Aptamers can mimic or block the function of various molecules involved in cancer progression [41].

Drug carriers: Their adaptable composition enables the attachment of therapeutic agents or delivery vehicles, resulting in higher drug concentration at the tumor location and enhanced therapeutic effectiveness [42].

Delivery of therapeutic oligonucleotides: Aptamers can be conjugated to oligonucleotide drugs like siRNAs for targeted delivery and modulation of gene expression within cancer cells [43].

Imaging agents: Aptamers can be conjugated to imaging agents for specific tumor diagnosis, allowing for earlier detection and more precise treatment planning [44]

Functional Characteristics and Stability

The functional characteristics and stability of aptamers are crucial for their effectiveness in clinical applications. Aptamers' stability, in vivo lifespan, and therapeutic efficiency can be improved by adding artificial nucleotides or encapsulating them in nanoparticles. Researches demonstrated that chemically modified aptamers exhibit increased resistance to enzymatic degradation and enhanced binding affinity, leading to improved therapeutic outcomes [25-27].

Aptamers demonstrate exceptional specificity and affinity towards their targets, with dissociation constants spanning from 10 pM to 10 nM. Their functional length typically ranges from 15 to 60 nucleotides, making them versatile for binding to various targets [45].

While inherently stable, aptamers can face challenges in vivo, including enzymatic degradation and rapid clearance from the body. Strategies to improve their stability include:

- Capping the ends of the aptamer with modified nucleotides
- Incorporating unnatural nucleotides or hydrocarbon linkers for enhanced stability
- Using L-enantiomers (mirror images) of natural nucleotides
- Employing locked nucleic acids (LNAs)
- Conjugation with polymers like polyethylene glycol (PEG) for improved half-life [46].

Methods for Isolation of Aptamers

In vitro selection is the standard procedure for isolating aptamers that exhibit high affinity and specificity for target molecules (SELEX).

Recent innovations in SELEX technology have further expanded its capabilities. Cell-SELEX allows for the selection of aptamers against complex targets on cell surfaces, while microfluidic-based SELEX has dramatically increased the speed and efficiency of the selection process. These advancements are pushing the boundaries of aptamer discovery and application in cancer therapy (Figure 1).

In Vitro Selection (SELEX):

SELEX, referred to as systematic evolution of ligands by exponential enrichment, is a repetitive procedure used to extract aptamers from a large collection of random single-stranded oligonucleotides (DNA or RNA). The initial library can consist of an immense number of distinct sequences, such as 1024 sequences each containing 40 randomly selected nucleotides, in order to guarantee a wide range of possible aptamers. The SELEX procedure has multiple essential stages [47, 48].

- Selection: The target molecule of interest is incubated with the random oligonucleotide library. Only oligonucleotides that possess a precise and strong ability to bind to the target will be kept.
- Washing: Unattached oligonucleotides are eliminated by means of washing procedures.
- Amplification: The bound oligonucleotides are extracted and amplified using PCR (for DNA) or reverse transcription PCR (RT-PCR) for RNA. This process increases the number of sequences with the ability to bind to the target.
- Iterative process: The stages of selecting, washing, and amplifying are done several times (usually 6-10) until aptamers that have a strong attraction and specificity for the target are gradually increased in quantity [47].

Following the completion of the final SELEX round, aptamers with a strong binding affinity are determined using methods such as sequencing. These candidate aptamers may undergo further optimization, such as size minimization to improve tissue penetration and nuclease stabilization using modified nucleotides (e.g., 2'-F pyrimidines, 2'-O-methyl nucleotides) to enhance in vivo stability [49].

Isolation of RNA Aptamers:

RNA aptamers require additional steps during SELEX compared to DNA aptamers:

Transcription: Following each selection round, the enriched pool of DNA (complementary DNA or cDNA from RNA) is transcribed back into RNA for the subsequent selection round.

Reverse Transcription: In the first round, RNA from the random library is converted into cDNA before PCR amplification.

This additional step allows for the incorporation of unnatural nucleotides (e.g., 2'-F pyrimidines, 2'-O-methyl nucleotides) during the transcription process. These modifications can introduce nuclease resistance to the RNA aptamers during selection, a significant advantage over DNA aptamers. For example, this method has yielded a fully 2'-O-methyl-modified anti-VEGF aptamer exhibiting an extended circulating half-life of 23 hours when conjugated to 40 kDa PEG [50]. In contrast, DNA aptamers lack a natural enzymatic machinery to incorporate unnatural bases during amplification and typically require post-SELEX chemical modifications for nuclease stabilization [51].

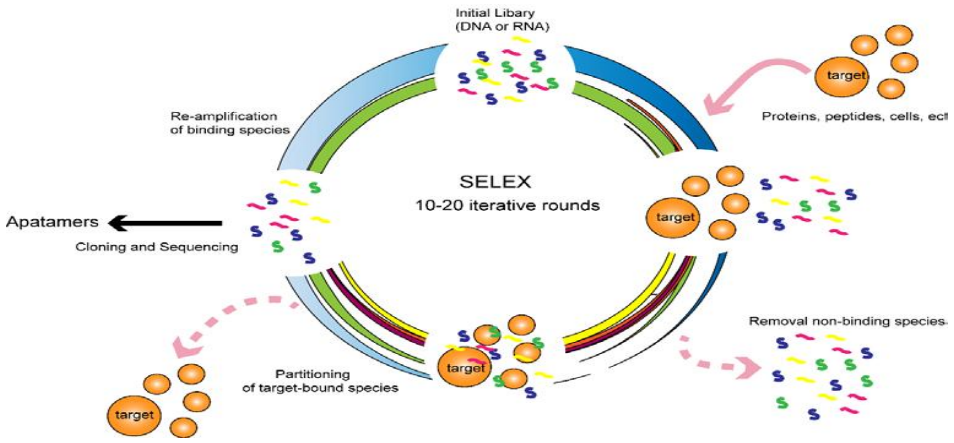

Figure 1: An explanatory diagram depicting the SELEX process. The initial collection of DNA or RNA oligonucleotides undergoes a series of repetitive processes including selection, partitioning, and

amplification. Following several iterations (usually 10-20), the concentrated collection of aptamers with strong binding capabilities is replicated and analyzed for their genetic sequence [48].

Aptamers: Generation, Selection, and Applications

Aptamers, short DNA or RNA molecules, offer specific and high-affinity binding to diverse molecular targets. Their unique three-dimensional structures enable precise recognition. The gold standard methodology for aptamer generation is SELEX, a technique established in 1990 (Figure 1) [52-54].

SELEX Process:

SELEX involves enriching sequences capable of binding a target from a random oligonucleotide library. This library, containing a 20–60 nucleotide-long random core, undergoes iterative selection rounds. The selected aptamers, influenced by the selection environment, exhibit specific binding to the target. The SELEX process allows high-throughput screening of billions of aptamers, identifying the most effective binders [55].

ssDNA vs. RNA Aptamers:

While ssDNA and RNA aptamers share functional similarities, differences emerge in their selection protocols. ssDNA aptamers are PCR-amplified and separated, while RNA aptamers require reverse transcription and in vitro transcription. RNA aptamers, offering greater conformational diversity, may enhance binding affinity but are more time-consuming to identify and less stable than ssDNA aptamers. The choice between them depends on specific requirements [56].

Enhancements in Aptamer Selection:

Advancements in oligonucleotide chemistry, synthesis methods, and technical analysis have improved aptamer discovery. Traditional SELEX protocols focused on purified recombinant proteins immobilized in solid-phase matrices. However, these methods may not fully represent the conformations of proteins found within living cells. To address this limitation, "live" SELEX approaches have been introduced:

Whole-cell SELEX: This approach utilizes intact cells as the target during the selection process. This allows aptamers to be selected based on their ability to bind to proteins in their native conformations, enhancing their potential efficacy in vivo [57].

Live-animal-based SELEX: This approach takes SELEX a step further by utilizing live animals. Here, aptamers are selected based on their ability to bind to target molecules within a living organism. This approach bridges the gap between in vitro selection and in vivo effects, allowing for the identification of aptamers that effectively target disease within a complex biological environment [58].

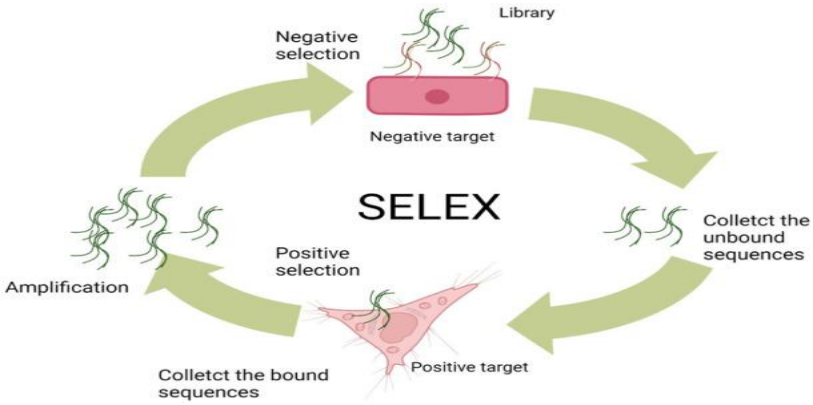

Figure 2: Schematic representation of the SELEX process. A library of random oligonucleotides is incubated with a negative target to remove sequences that bind to it (negative selection). The remaining sequences are then incubated with the positive target and the bound sequences are collected. These sequences are amplified, and the process is repeated for several rounds. The final enriched pool is then sequenced to identify the aptamers that bind specifically to the positive target [59].

Types of Nanoparticles for Aptamer Conjugation

Aptamer-drug conjugates (ADCs) represent a novel class of targeted therapy, offering specificity and reduced toxicity. A comprehensive study by Vandghanooni et al. (2019) showcased the potential of ADCs in precisely delivering chemotherapeutics to cancer cells, significantly reducing off-target effects and improving therapeutic indices [60]. This research highlights the potential of ADCs to transform cancer treatment by combining the targeted delivery of aptamers with the therapeutic efficacy of conventional drugs.

The choice of nanoparticles for aptamer conjugation significantly affects delivery properties. Commonly used types include:

Polymeric nanoparticles: These versatile carriers enable controlled drug release, allowing for sustained therapeutic effects. Additionally, polymeric nanoparticles can be tailored with specific targeting functionalities through surface modifications, enhancing their ability to home in on cancer cells and improving drug delivery efficiency **[61].**

Liposomes: These biocompatible vesicles are another powerful tool for aptamer conjugation. Liposomes can encapsulate a wide range of drugs, including both water-soluble and water-insoluble molecules, protecting them from degradation and enhancing their stability in the body. Moreover, liposomes exhibit improved biocompatibility, minimizing potential side effects **[62].**

Engineering Considerations for Nanoparticle-Aptamer Conjugation

Engineering nanoparticles for aptamer conjugation is crucial for enhancing drug delivery to cancer cells. Recent advancements highlight innovative strategies in nanoparticle design that improve cellular uptake, biodistribution, and therapeutic efficacy [30]. These strategies involve optimizing nanoparticle size, surface charge, and conjugation techniques, which are essential for the successful application of nanoparticle-aptamer complexes in cancer therapy (Figure 3).

he engineering of nanoparticles for aptamer conjugation is a nuanced process that requires consideration of particle size, surface charge, and biocompatibility to optimize delivery

efficacy and minimize off-target effects. This precision ensures that therapeutic agents reach the intended cancer cells. ADCs offer several advantages:

- **Targeted delivery:** Aptamers specifically bind to cancer cells, guiding the conjugated drug directly to the tumor site. This targeted approach minimizes exposure of healthy tissues to the drug, reducing potential side effects [29].
- **Enhanced drug efficacy:** Targeted delivery via aptamers allows for higher drug concentrations at the tumor site compared to traditional therapies, leading to improved therapeutic efficacy and potentially lower drug doses [63].

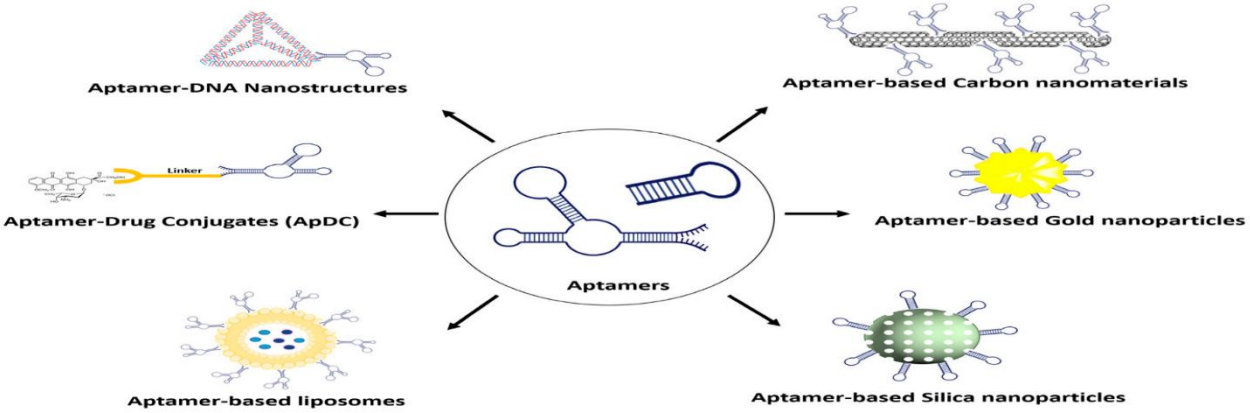

Figure 3: A variety of aptamer conjugates for targeted drug delivery systems. This figure illustrates the different strategies for conjugating aptamers to nanoparticles, including covalent and noncovalent methods.

Nanoparticles offer a versatile platform for delivering drugs [64]. Key considerations for engineering nanoparticles for optimal aptamer conjugation:

Ideal nanoparticle characteristics:

Size: Nanoparticles with a size range of 10-200 nm are ideal for effective tumor penetration. This size allows them to navigate the complex tumor microenvironment and reach cancer cells [65].

Drug loading capacity: High drug loading capacity is crucial for maximizing the therapeutic potential of the conjugate. Nanoparticles should be designed to efficiently encapsulate sufficient drug quantities for effective treatment.

Blood clearance: Nanoparticles with slower blood clearance times allow for extended circulation in the body. This extended circulation time increases the probability of reaching and accumulating within the tumor site [66].

Targeted delivery: Aptamer conjugation is a key strategy for achieving targeted delivery. Aptamers specifically bind to cancer cells, guiding the nanoparticle-drug conjugate to the desired location.

Nanoparticle engineering considerations:

Biomaterial selection: The choice of biomaterial for nanoparticle fabrication significantly impacts drug encapsulation and release properties. For instance, polymers like PLGA (poly(lactic-co-glycolic acid)) and PLA (polylactic acid) are commonly used due to their biocompatibility and tunable degradation profiles [67, 68].

Size control: Precise control over nanoparticle size is essential. This can be achieved by adjusting factors like solvent ratios and polymer concentrations during the nanoparticle fabrication process [68].

Surface modification: Surface modification plays a critical role in biodistribution, targeting, and stability of nanoparticles. Strategies like PEGylation (attachment of polyethylene glycol) can enhance blood circulation time by reducing unwanted interactions with blood components. Additionally, surface modifications can be employed to incorporate aptamers for targeted delivery [39, 67].

Size and Biodistribution of Nanoparticles:

Smaller nanoparticles (<150 nm) exhibit several advantages. They demonstrate superior tumor penetration due to their ability to navigate the extracellular matrix more effectively. Additionally, smaller nanoparticles are less likely to be taken up by macrophages, immune cells that can clear nanoparticles from circulation [69, 70].

Charge of Nanoparticles:

The surface charge of nanoparticles significantly impacts their behavior in the body. Factors like opsonization (recognition and engulfment by immune cells) and biodistribution are influenced by surface charge [71].

For optimal aptamer conjugation, neutral or negatively charged nanoparticles are generally preferred. This charge profile minimizes unwanted interactions with blood components and enhances circulation time [70].

Conjugation Strategies for Nanoparticle-Aptamer

The successful conjugation of aptamers and nanoparticles hinges on efficient conjugation strategies. Two main approaches are employed:

Covalent conjugation: This strategy involves forming a direct chemical bond between the aptamer and the nanoparticle. Common covalent conjugation methods utilize reactions between functional groups like succinimidyl esters and amines. Covalent conjugation offers several advantages. It creates a stable linkage between the aptamer and nanoparticle, ensuring they remain tethered throughout delivery. Additionally, covalent conjugation avoids the introduction of unnecessary biological components, potentially minimizing unwanted immune reactions [72].

Noncovalent conjugation: This approach relies on weaker, reversible interactions to link aptamers to nanoparticles. A common noncovalent strategy utilizes the high-affinity interaction between biotin and streptavidin. By attaching biotin to the aptamer and streptavidin to the nanoparticle surface, a strong, yet reversible, bond is formed [72]. Noncovalent conjugation can offer advantages like tunability and the potential for triggered release under specific conditions. However, careful design is necessary to ensure sufficient stability during circulation and delivery to the target site.

Aptamer-Drug Conjugates for Targeted Cancer Therapy

Aptamer-drug conjugates (ADCs) represent a revolutionary approach in targeted cancer therapy. They combine the targeting prowess of aptamers with the therapeutic potential of drugs (Figure 4 and 5), offering several advantages:

Targeted delivery: Aptamers act as homing beacons, specifically binding to cancer cells. This targeted approach directs the conjugated drug to the tumor site, minimizing exposure of healthy tissues to the therapeutic agent. This targeted approach can significantly reduce the side effects associated with conventional cancer therapies [29, 63].

Enhanced drug efficacy: By delivering drugs directly to cancer cells, ADCs can achieve higher intratumorally drug concentrations compared to traditional therapies. This targeted delivery can lead to improved treatment efficacy and potentially allows for lower drug doses, reducing the risk of systemic side effects.

Design versatility: Aptamers can be conjugated to a broad range of therapeutic agents, including small molecule drugs, chemotherapeutics, and even siRNA (short interfering RNA) for gene silencing. This versatility allows for the development of ADCs tailored to target specific cancers and therapeutic needs [28].

The design of ADCs goes beyond simply linking an aptamer to a drug. Several key considerations influence their efficacy:

Site-selective functionalization: Precise attachment of the drug molecule to a designated location on the aptamer is crucial. This ensures consistent drug orientation and facilitates optimal therapeutic activity upon release [31].

Cleavable linkers: The linker molecule connecting the aptamer and drug plays a vital role. Ideally, the linker should be stable during circulation but cleavable under specific conditions at the tumor site to trigger drug release. Disulfide linkages are a common example, as they can be cleaved by the reducing environment present within tumor cells [32].

Controlled release: The size and properties of the ADC, including the linker chemistry, can influence the rate of drug release. By carefully designing the ADC, researchers can achieve controlled drug release profiles, optimizing therapeutic efficacy and minimizing potential side effects [33].

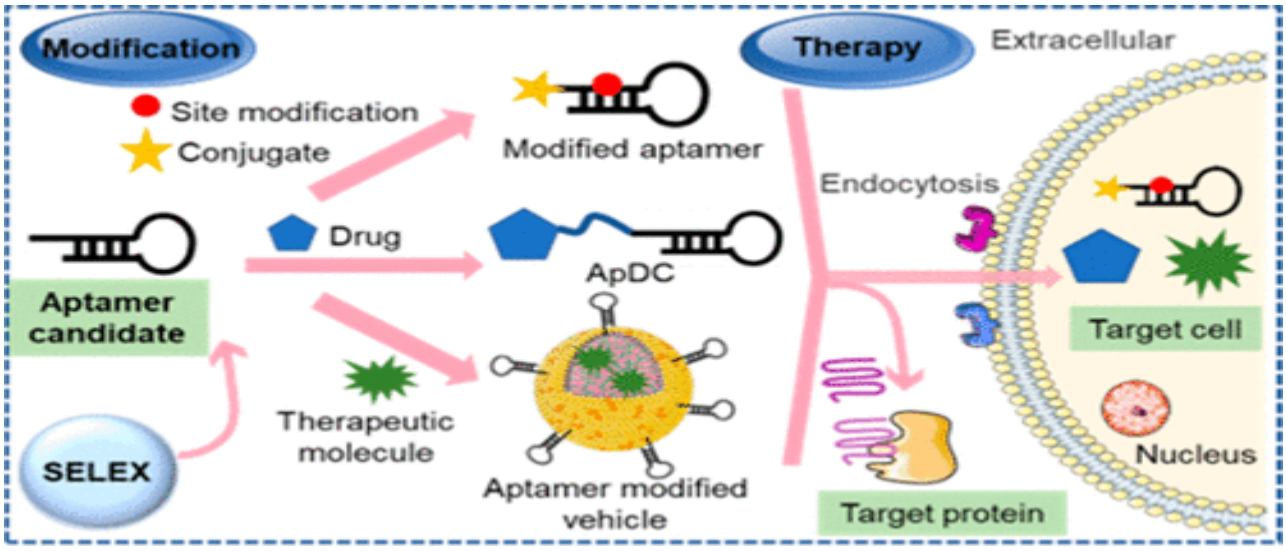

Figure 4: The figure illustrates the application of aptamers in targeted therapy through various modifications and delivery strategies. The process begins with SELEX for the selection of aptamer candidates, followed by modifications such as site-specific labeling (red circle) and conjugation (yellow star) with drugs or therapeutic molecules.

Aptamers in Nanoparticle-Based Cancer Therapeutics

The potential of aptamers is significantly amplified when combined with nanoparticles for targeted drug delivery in cancer therapy. Aptamers conjugated to nanoparticles can specifically target cancer cells while minimizing off-target effects, reducing potential side effects [34].

Applications of Aptamers in Nanoparticle-Based Cancer Delivery:

Several aptamers have been identified with therapeutic potential in various cancers by targeting specific proteins expressed in different cancer types. Table 1 provides some examples of aptamers under investigation for targeted cancer therapy using nanoparticle conjugates.

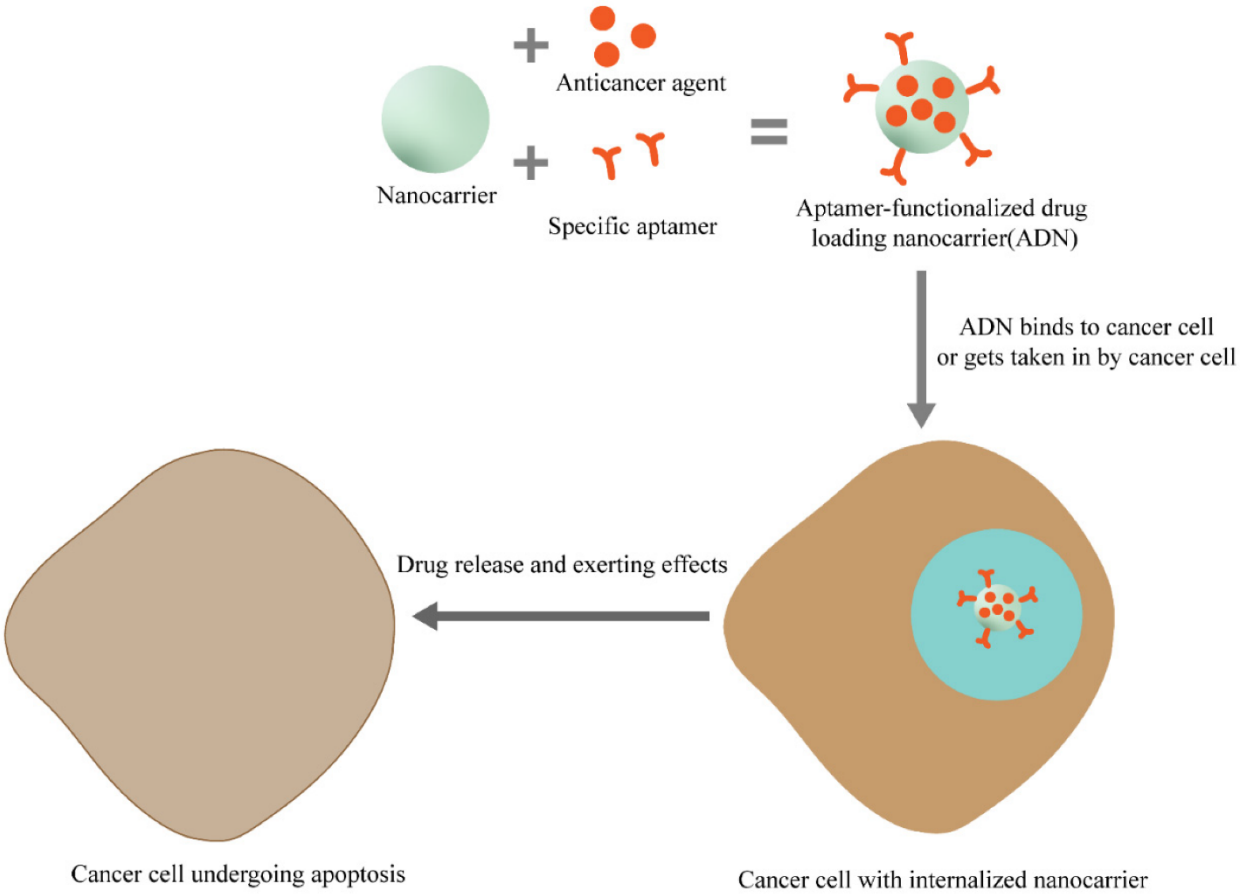

Figure 5: Aptamer-functionalized nanoparticles acting on a cancer cell. This figure depicts how nanoparticles, functionalized with aptamers, can selectively target and deliver therapeutic agents to cancer cells.

Clinical Applications of Aptamer-Based Therapies

Several aptamer-based therapies are currently in various stages of clinical trials for cancer treatment. For example, AS1411, a DNA aptamer targeting nucleolin, has shown promise in phase II trials for acute myeloid leukemia. Another aptamer, NOX-A12, targeting CXCL12, is being evaluated in combination with checkpoint inhibitors for colorectal and pancreatic cancer.

However, regulatory challenges specific to aptamer-based therapeutics remain. These include standardizing production processes, ensuring batch-to-batch consistency, and developing appropriate safety and efficacy testing protocols. Overcoming these hurdles will be crucial for the widespread clinical adoption of aptamer-based cancer therapies (Table 1).

Table 1: Examples of Aptamers for Targeted Delivery in Cancer Therapy

Aptamer	Full Name	Function	Cancers Associated With	References
A30	RNA aptamer	Inhibits HER-3 function	Breast cancer (HER-2 overexpression)	[73]
Apt-avb3		Mediates angiogenesis and tumor growth	Not specified	[74]
ARGO100 (AS1411)	DNA aptamer	Modulates NF-kB pathways	Leukemia, gastric, and breast cancers	[75]
CD134 (OX40) aptamer 9.8	RNA aptamer	Stimulates immune response	Melanoma	[76]
CD137 (4-1BB) aptamer PSMA-4-1BB	RNA aptamer	Stimulates immune response	Prostate cancer	[77]
CD44 aptamer TA1	DNA aptamer	Modulates CD44-mediated processes	Breast, prostate, and cancer stem cells	[78]
Clone 5	RNA aptamer	Binds to sLex	Not specified	[79]
D60	RNA aptamer	Inhibits CTLA-4 function	Not specified	[80]
EpCAM aptamer SYL3	DNA aptamer	Regulates gene expression	Bladder, breast, colon, lung, ovarian, pancreatic, and prostate cancers	[81]
EpCAM aptamer EpDT3-DY647	RNA aptamer	Not specified	Not specified	[82, 83]
EpCAM aptamer (Apt/RNA)	RNA aptamer	Not specified	Not specified	[84]

HB5/DNA	DNA aptamer	Inhibits tumorigenic signaling	Breast, gastric, lung, colorectal, esophageal, and ovarian cancers	[85]
HER2 aptamer (Apt/DNA)	DNA aptamer	Inhibits tumorigenic signaling	Not specified	[85, 86]
MUC1 aptamer S2.1/DNA	DNA aptamer	Prevents cancer cell invasion	Ovarian, breast, lung, pancreatic cancers, and multiple myeloma	[87, 88]
MUC1 aptamer (Apt/DNA)	DNA aptamer	Prevents cancer cell invasion	Not specified	[89]
NF-kB aptamer (Apt/RNA)	RNA aptamer	Modulates NF-kB pathways	Cervical, prostate, lung, and breast cancers	[82]
VEGF (vascular endothelial growth factor NX-191	RNA aptamer	Targets VEGF pathways	Breast, brain, lung, colon, gastric, pancreatic, melanoma, myeloid, and leukemia	[90]
VEGF NX-213	RNA aptamer	Targets VEGF pathways	Breast, brain, lung, colon, gastric, pancreatic, melanoma, myeloid, and leukemia	[91]
PD-1 aptamer MP5/MP7	DNA aptamer	Modulates PD-1-associated immune response	Colon cancer, carcinoma	[78]
PDGF aptamer 36t	DNA aptamer	Targets PDGF pathways	Ovarian, breast, thyroid, cervical, and lung cancers	[92]
PSMA aptamer xPSM-A9	RNA aptamer	Inhibits PSMA function	Prostate cancer	[92]
PSMA aptamer xPSM-A10	RNA aptamer	Inhibits PSMA function	Prostate cancer	[93]
PSMA aptamer (Apt/DNA)	DNA aptamer	Not specified	Prostate cancer	[94, 95]
Sialyl Lewis X (sLex) aptamer Clone 5	RNA aptamer	Inhibits sLex/selectin-mediated adhesion	Not specified	[79]

Tenascin-C aptamer TTA1		Not specified	Not specified	[96]
Vap7/V7t1	DNA aptamer	Targets VEGF pathways	Not specified	[97]

Future Directions

Aptamer-drug conjugates (ADCs) hold significant promise for targeted cancer therapy. Ongoing research is focused on improving several aspects of ADCs:

Delivery Efficiency: Enhancing the efficiency of ADC delivery to tumor sites is crucial for maximizing therapeutic efficacy.

Specificity: Further refinement of aptamer targeting ensures precise delivery of the conjugated drug to cancer cells, minimizing off-target effects on healthy tissues.

Therapeutic Outcomes: Optimizing the therapeutic potential of ADCs involves strategies to improve drug release profiles and enhance overall treatment efficacy.

While ADCs offer exciting possibilities, some challenges remain:

Limited Toxicological Information: Limited toxicological data on aptamers in humans necessitates further investigation to ensure their safety.

Clinical Translation Infrastructure: Developing the infrastructure and protocols necessary for widespread clinical translation of aptamers is an ongoing effort.

Overall, aptamer-drug conjugates represent a promising approach for targeted cancer therapy with unique advantages over traditional antibody-based methods. Continued research holds the potential to overcome current hurdles and establish aptamers as next-generation therapeutics for various cancers [98, 99].

Emerging trends in aptamer-based cancer therapies include their combination with immunotherapy approaches. For instance, aptamers targeting immune checkpoint proteins like PD-1 or CTLA-4 could enhance the efficacy of existing immunotherapies. Additionally, aptamers show promise in overcoming drug resistance by targeting specific resistance mechanisms or by providing alternative delivery routes for established drugs.

Another exciting avenue is the development of aptamer-based theranostic agents, which combine diagnostic and therapeutic functions. These could allow for real-time monitoring of drug delivery and therapeutic response, paving the way for more personalized cancer treatment strategies.

Conclusion: A Future of Targeted Cancer Therapy with Aptamers

This manuscript has explored the exciting potential of aptamers for targeted cancer therapy. Aptamers, single-stranded nucleic acid molecules, offer several advantages over traditional antibodies, including smaller size, higher tissue permeability, and reduced immunogenicity. These properties make them ideal candidates for delivering therapeutic agents directly to cancer cells with greater precision.

The development of Aptamer-Drug Conjugates (ADCs) represents a significant leap forward in targeted cancer therapy. By combining aptamers with various therapeutic agents, such as

small molecules, proteins, and nucleic acids, ADCs can deliver a precise payload to cancer cells while minimizing damage to healthy tissues.

Nanoparticles offer a versatile platform for delivering aptamer-based therapeutics. By engineering these nanoparticles with specific characteristics like size, surface charge, and biodegradability, researchers can optimize drug delivery and enhance therapeutic efficacy. Surface modification plays a crucial role in this process, enabling the attachment of aptamers and preventing unwanted interactions with healthy tissues.

In conclusion, aptamer-based nanoparticle bioconjugates represent a significant leap forward in targeted cancer therapy. Their unique properties - including high specificity, low immunogenicity, and ease of modification - position them as powerful tools for precise drug delivery. As we overcome challenges related to in vivo stability and optimize delivery strategies, these bioconjugates have the potential to dramatically improve cancer treatment outcomes. The integration of aptamers with emerging technologies like immunotherapy and personalized medicine heralds a new era in cancer treatment, where therapies can be tailored with unprecedented precision to individual patients and tumor types.

Future Outlook

Despite the promising preclinical data, aptamer-based therapies are still in their early stages of development. Challenges remain, including limited toxicological information in humans and the need for infrastructure for widespread clinical translation. However, ongoing research is actively addressing these challenges:

Improved Delivery Efficiency: Efforts are underway to enhance the efficiency of ADC delivery to tumor sites, maximizing therapeutic efficacy.

Enhanced Specificity: Further refinement of aptamer targeting ensures precise delivery of the conjugated drug to cancer cells, minimizing off-target effects on healthy tissues.

Optimized Therapeutic Outcomes: Research is focused on optimizing the therapeutic potential of ADCs, including strategies to improve drug release profiles and enhance overall treatment efficacy.

The field of aptamer-based therapies is rapidly advancing, with ongoing clinical trials evaluating their efficacy in various cancers. Additionally, research is exploring strategies to further improve aptamer targeting, drug release profiles, and overall therapeutic efficacy.

In conclusion, aptamers hold immense promise for revolutionizing targeted cancer therapy due to their unique properties and versatility. Future research should prioritize enhancing the delivery efficiency, specificity, and therapeutic outcomes of aptamer-drug conjugates. Continued advancements in this field could establish aptamers as next-generation therapeutics for various cancers, offering hope for more effective and selective treatments.

By implementing these improvements, the review will be more concise, clear, and focused, providing a stronger foundation for understanding the advancements in aptamer-based nanoparticle bioconjugates for targeted cancer therapy.

REFERENCES

1. Pierpont TM, Limper CB, Richards KL. Past, present, and future of rituximab—the world's first oncology monoclonal antibody therapy. Front Oncol. 2018;8:163. https://doi.org/10.3389/fonc.2018.00163.
2. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. N Engl J Med. 2001;344(11):783-92. https://doi.org/10.1056/NEJM200103153441101.
3. Ferrara N, Hillan KJ, Gerber H-P, Novotny W. Discovery and development of bevacizumab, an anti-VEGF antibody for treating cancer. Nature reviews Drug discovery. 2004;3(5):391-400. https://doi.org/10.1038/nrd1381.
4. Van Cutsem E, Köhne C-H, Hitre E, Zaluski J, Chang Chien C-R, Makhson A, et al. Cetuximab and chemotherapy as initial treatment for metastatic colorectal cancer. N Engl J Med. 2009;360(14):1408-17. https://doi.org/10.1056/NEJMoa0805019.
5. Alpdogan O, Kartan S, Johnson W, Sokol K, Porcu P. Systemic therapy of cutaneous T-cell lymphoma (CTCL). Chinese Journal of Clinical Oncology. 2019;8(1):10.
6. Quinteros DA, Bermúdez JM, Ravetti S, Cid A, Allemandi DA, Palma SD. Therapeutic use of monoclonal antibodies: General aspects and challenges for drug delivery. Nanostructures for Drug Delivery: Elsevier; 2017. p. 807-33.
7. Mitra S, Tomar PC. Hybridoma technology; advancements, clinical significance, and future aspects. Journal of Genetic Engineering and Biotechnology. 2021;19(1):1-12.
8. Tuerk C, Gold L. Systematic evolution of ligands by exponential enrichment: RNA ligands to bacteriophage T4 DNA polymerase. Science. 1990;249(4968):505-10.
9. Ni X, Castanares M, Mukherjee A, Lupold SE. Nucleic acid aptamers: clinical applications and promising new horizons. Curr Med Chem. 2011;18(27):4206-14.
10. Gold L, Janjic N, Jarvis T, Schneider D, Walker JJ, Wilcox SK, et al. Aptamers and the RNA world, past and present. Cold Spring Harb Perspect Biol. 2012;4(3). https://doi.org/10.1101/cshperspect.a003582.
11. Marshall ML, Wagstaff KM. Internalized functional DNA aptamers as alternative cancer therapies. Front Pharmacol. 2020;11:1115.
12. Wang X, Zhou Q, Li X, Gan X, Liu P, Feng X, et al. Insights into Aptamer-Drug Delivery Systems against Prostate Cancer. Molecules. 2022;27(11). https://doi.org/10.3390/molecules27113446.
13. Omid C. Farokhzad SJ, Robert Langer. Aptamers and Cancer Nanotechnology. In: Amiji MM, editor. Nanotechnology for cancer therapy. 1 ed. New York, USA: CRC press; 2006. p. 289-313.
14. Chehelgerdi M, Chehelgerdi M, Allela OQB, Pecho RDC, Jayasankar N, Rao DP, et al. Progressing nanotechnology to improve targeted cancer treatment: overcoming hurdles in its clinical implementation. Mol Cancer. 2023;22(1):169. https://doi.org/10.1186/s12943-023-01865-0.
15. Dausse E, Taouji S, Evadé L, Di Primo C, Chevet E, Toulmé J-J. HAPIscreen, a method for high-throughput aptamer identification. Journal of Nanobiotechnology. 2011;9(1):25. https://doi.org/10.1186/1477-3155-9-25.
16. Kumar Kulabhusan P, Hussain B, Yüce M. Current perspectives on aptamers as diagnostic tools and therapeutic agents. Pharmaceutics. 2020;12(7):646.
17. Röthlisberger P, Hollenstein M. Aptamer chemistry. Advanced Drug Delivery Reviews. 2018;134:3-21. https://doi.org/https://doi.org/10.1016/j.addr.2018.04.007.

18. Shraim AaS, Abdel Majeed BA, Al-Binni MA, Hunaiti A. Therapeutic Potential of Aptamer–Protein Interactions. ACS Pharmacology & Translational Science. 2022;5(12):1211-27.
19. Zhou G, Wilson G, Hebbard L, Duan W, Liddle C, George J, et al. Aptamers: A promising chemical antibody for cancer therapy. Oncotarget. 2016;7(12):13446.
20. Mo T, Liu X, Luo Y, Zhong L, Zhang Z, Li T, et al. Aptamer-based biosensors and application in tumor theranostics. Cancer Sci. 2022;113(1):7-16. https://doi.org/10.1111/cas.15194.
21. Rabiee N, Chen S, Ahmadi S, Veedu RN. Aptamer-engineered (nano)materials for theranostic applications. Theranostics. 2023;13(15):5183-206. https://doi.org/10.7150/thno.85419.
22. Odeh F, Nsairat H, Alshaer W, Ismail MA, Esawi E, Qaqish B, et al. Aptamers Chemistry: Chemical Modifications and Conjugation Strategies. Molecules. 2019;25(1). https://doi.org/10.3390/molecules25010003.
23. Sun H, Zhu X, Lu PY, Rosato RR, Tan W, Zu Y. Oligonucleotide Aptamers: New Tools for Targeted Cancer Therapy. Molecular Therapy - Nucleic Acids. 2014;3:e182. https://doi.org/https://doi.org/10.1038/mtna.2014.32.
24. Szymanowski W, Szymanowska A, Bielawska A, Lopez-Berestein G, Rodriguez-Aguayo C, Amero P. Aptamers as potential therapeutic tools for ovarian cancer: Advancements and challenges. Cancers (Basel). 2023;15(21):5300.
25. Chen Z, Luo H, Gubu A, Yu S, Zhang H, Dai H, et al. Chemically modified aptamers for improving binding affinity to the target proteins via enhanced non-covalent bonding. Frontiers in Cell and Developmental Biology. 2023;11:1091809.
26. Elskens JP, Elskens JM, Madder A. Chemical modification of aptamers for increased binding affinity in diagnostic applications: Current status and future prospects. Int J Mol Sci. 2020;21(12):4522.
27. Nimjee SM, Sullenger BA. Therapeutic Aptamers: Evolving to Find their Clinical Niche. Curr Med Chem. 2020;27(25):4181-93. https://doi.org/10.2174/0929867326666191001125101.
28. Chandola C, Neerathilingam M. Aptamers for targeted delivery: current challenges and future opportunities. In: Rajeev K. Tyagi NG, Rahul Shukla and Prakash Singh Bisen, editor. Role of novel drug delivery vehicles in nanobiomedicine2019. p. 1-22.
29. Liu B, Liu J, Hu X, Xiang W, Hou W, Li C, et al. Recent advances in aptamer-based therapeutic strategies for targeting cancer stem cells. Mater Today Bio. 2023;19:100605. https://doi.org/10.1016/j.mtbio.2023.100605.
30. Mahmoudian F, Ahmari A, Shabani S, Sadeghi B, Fahimirad S, Fattahi F. Aptamers as an approach to targeted cancer therapy. Cancer Cell Int. 2024;24(1):108. https://doi.org/10.1186/s12935-024-03295-4.
31. Kim DH, Seo JM, Shin KJ, Yang SG. Design and clinical developments of aptamer-drug conjugates for targeted cancer therapy. Biomater Res. 2021;25(1):42. https://doi.org/10.1186/s40824-021-00244-4.
32. Sheyi R, de la Torre BG, Albericio F. Linkers: An assurance for controlled delivery of antibody-drug conjugate. Pharmaceutics. 2022;14(2):396.
33. Thang NH, Chien TB, Cuong DX. Polymer-based hydrogels applied in drug delivery: An overview. Gels. 2023;9(7):523.
34. Wei Z, Zhou Y, Wang R, Wang J, Chen Z. Aptamers as Smart Ligands for Targeted Drug Delivery in Cancer Therapy. Pharmaceutics. 2022;14(12). https://doi.org/10.3390/pharmaceutics14122561.

35. Adepu S, Ramakrishna S. Controlled drug delivery systems: current status and future directions. Molecules. 2021;26(19):5905.
36. Zhang T, Wang Z. Monoclonal Antibody Development for Cancer Treatment Using the Phage Display Library Platform. Biologics. 2024;4(1):55-74.
37. Chehelgerdi M, Chehelgerdi M, Allela OQB, Pecho RDC, Jayasankar N, Rao DP, et al. Progressing nanotechnology to improve targeted cancer treatment: overcoming hurdles in its clinical implementation. Mol Cancer. 2023;22(1):169. https://doi.org/10.1186/s12943-023-01865-0.
38. Lu R-M, Hwang Y-C, Liu IJ, Lee C-C, Tsai H-Z, Li H-J, et al. Development of therapeutic antibodies for the treatment of diseases. J Biomed Sci. 2020;27(1):1. https://doi.org/10.1186/s12929-019-0592-z.
39. Zhu L, Yang J, Ma Y, Zhu X, Zhang C. Aptamers Entirely Built from Therapeutic Nucleoside Analogues for Targeted Cancer Therapy. J Am Chem Soc. 2022;144(4):1493-7. https://doi.org/10.1021/jacs.1c09574.
40. Zhou J, Rossi J. Aptamers as targeted therapeutics: current potential and challenges. Nat Rev Drug Discov. 2017;16(3):181-202. https://doi.org/10.1038/nrd.2016.199.
41. Amero P, Khatua S, Rodriguez-Aguayo C, Lopez-Berestein G. Aptamers: Novel therapeutics and potential role in neuro-oncology. Cancers (Basel). 2020;12(10):2889.
42. Guan B, Zhang X. Aptamers as Versatile Ligands for Biomedical and Pharmaceutical Applications. Int J Nanomedicine. 2020;15:1059-71. https://doi.org/10.2147/ijn.S237544.
43. Allemailem KS, Almatroudi A, Alsahli MA, Basfar GT, Alrumaihi F, Rahmani AH, et al. Recent advances in understanding oligonucleotide aptamers and their applications as therapeutic agents. 3 Biotech. 2020;10:1-20.
44. Xuan W, Peng Y, Deng Z, Peng T, Kuai H, Li Y, et al. A basic insight into aptamer-drug conjugates (ApDCs). Biomaterials. 2018;182:216-26. https://doi.org/10.1016/j.biomaterials.2018.08.021.
45. Wang B, Kobeissy F, Golpich M, Cai G, Li X, Abedi R, et al. Aptamer Technologies in Neuroscience, Neuro-Diagnostics and Neuro-Medicine Development. Molecules. 2024;29(5):1124.
46. Shigdar S, Macdonald J, O'Connor M, Wang T, Xiang D, Al. Shamaileh H, et al. Aptamers as theranostic agents: modifications, serum stability and functionalisation. Sensors. 2013;13(10):13624-37.
47. Bayat P, Nosrati R, Alibolandi M, Rafatpanah H, Abnous K, Khedri M, et al. SELEX methods on the road to protein targeting with nucleic acid aptamers. Biochimie. 2018;154:132-55.
48. Zhuo Z, Yu Y, Wang M, Li J, Zhang Z, Liu J, et al. Recent advances in SELEX technology and aptamer applications in biomedicine. Int J Mol Sci. 2017;18(10):2142. https://doi.org/10.3390/ijms18102142.
49. Xu Y, Jiang X, Zhou Y, Ma M, Wang M, Ying B. Systematic evolution of ligands by exponential enrichment technologies and aptamer-based applications: Recent progress and challenges in precision medicine of infectious diseases. Frontiers in Bioengineering and Biotechnology. 2021;9:704077.
50. Szeto K, Latulippe DR, Ozer A, Pagano JM, White BS, Shalloway D, et al. Rapid-SELEX for RNA aptamers. PLoS One. 2013;8(12):e82667.
51. Umar MI, Chan C-Y, Kwok CK. Development of RNA G-quadruplex (rG4)-targeting l-RNA aptamers by rG4-SELEX. Nature Protocols. 2022;17(6):1385-414. https://doi.org/10.1038/s41596-022-00679-6.

52. Kong HY, Byun J. Nucleic Acid aptamers: new methods for selection, stabilization, and application in biomedical science. Biomol Ther (Seoul). 2013;21(6):423-34. https://doi.org/10.4062/biomolther.2013.085.
53. El-Husseini DM, Sayour AE, Melzer F, Mohamed MF, Neubauer H, Tammam RH. Generation and Selection of Specific Aptamers Targeting Brucella Species through an Enhanced Cell-SELEX Methodology. Int J Mol Sci. 2022;23(11):6131.
54. Amundarain A, Pastor F, Prósper F, Agirre X. Aptamers, a New Therapeutic Opportunity for the Treatment of Multiple Myeloma. Cancers (Basel). 2022;14(21):5471.
55. Komarova N, Kuznetsov A. Inside the black box: what makes SELEX better? Molecules. 2019;24(19):3598.
56. Chang ZY, Alhamami FAMS, Chin KL. Aptamer-based strategies to address challenges in COVID-19 diagnosis and treatments. Interdiscip Perspect Infect Dis. 2023;2023.
57. Kohlberger M, Gadermaier G. SELEX: Critical factors and optimization strategies for successful aptamer selection. Biotechnol Appl Biochem. 2022;69(5):1771-92. https://doi.org/10.1002/bab.2244.
58. Zhang Y, Lai BS, Juhas M. Recent advances in aptamer discovery and applications. Molecules. 2019;24(5):941.
59. Wu X, Shaikh AB, Yu Y, Li Y, Ni S, Lu A, et al. Potential diagnostic and therapeutic applications of oligonucleotide aptamers in breast cancer. Int J Mol Sci. 2017;18(9):1851.
60. Vandghanooni S, Eskandani M, Barar J, Omidi Y. Aptamedicine: a new treatment modality in personalized cancer therapy. Bioimpacts. 2019;9(2):67-70. https://doi.org/10.15171/bi.2019.09.
61. Fu Z, Xiang J. Aptamer-Functionalized Nanoparticles in Targeted Delivery and Cancer Therapy. Int J Mol Sci. 2020;21(23). https://doi.org/10.3390/ijms21239123.
62. Wong K-Y, Wong M-S, Liu J. Aptamer-functionalized liposomes for drug delivery. Biomedical Journal. 2023:100685. https://doi.org/https://doi.org/10.1016/j.bj.2023.100685.
63. Mohammadinejad A, Gaman LE, Aleyaghoob G, Gaceu L, Mohajeri SA, Moga MA, et al. Aptamer-Based Targeting of Cancer: A Powerful Tool for Diagnostic and Therapeutic Aims. Biosensors (Basel). 2024;14(2). https://doi.org/10.3390/bios14020078.
64. Alhamhoom Y, As Sobeai HM, Alsanea S, Alhoshani A. Aptamer-based therapy for targeting key mediators of cancer metastasis (Review). Int J Oncol. 2022;60(6). https://doi.org/10.3892/ijo.2022.5355.
65. Chen Y, Lin JS. The application of aptamer in apoptosis. Biochimie. 2017;132:1-8. https://doi.org/10.1016/j.biochi.2016.10.008.
66. Xie S, Ai L, Cui C, Fu T, Cheng X, Qu F, et al. Functional Aptamer-Embedded Nanomaterials for Diagnostics and Therapeutics. ACS Appl Mater Interfaces. 2021;13(8):9542-60. https://doi.org/10.1021/acsami.0c19562.
67. Del Amo L, Cano A, Ettcheto M, Souto EB, Espina M, Camins A, et al. Surface Functionalization of PLGA Nanoparticles to Increase Transport across the BBB for Alzheimer's Disease. Applied Sciences. 2021;11(9):4305.
68. Guo J, Gao X, Su L, Xia H, Gu G, Pang Z, et al. Aptamer-functionalized PEG–PLGA nanoparticles for enhanced anti-glioma drug delivery. Biomaterials. 2011;32(31):8010-20.
69. Dhar S, Gu FX, Langer R, Farokhzad OC, Lippard SJ. Targeted delivery of cisplatin to prostate cancer cells by aptamer functionalized Pt (IV) prodrug-PLGA–PEG nanoparticles. Proc Natl Acad Sci U S A. 2008;105(45):17356-61.

70. Li Y, Zhao J, Xue Z, Tsang C, Qiao X, Dong L, et al. Aptamer nucleotide analog drug conjugates in the targeting therapy of cancers. Front Cell Dev Biol. 2022;10:1053984. https://doi.org/10.3389/fcell.2022.1053984.
71. Zhao B, Chen S, Hong Y, Jia L, Zhou Y, He X, et al. Research Progress of Conjugated Nanomedicine for Cancer Treatment. Pharmaceutics. 2022;14(7). https://doi.org/10.3390/pharmaceutics14071522.
72. Dou XQ, Fu J, Song HF. [Advances in the study of aptamer-based drug for targeting therapy]. Yao Xue Xue Bao. 2016;51(7):1068-76.
73. Chen C-hB, Chernis GA, Hoang VQ, Landgraf R. Inhibition of heregulin signaling by an aptamer that preferentially binds to the oligomeric form of human epidermal growth factor receptor-3. Proc Natl Acad Sci U S A. 2003;100(16):9226-31.
74. Hori S-i, Herrera A, Rossi JJ, Zhou J. Current advances in aptamers for cancer diagnosis and therapy. Cancers (Basel). 2018;10(1):9.
75. Soundararajan S, Chen W, Spicer EK, Courtenay-Luck N, Fernandes DJ. The nucleolin targeting aptamer AS1411 destabilizes Bcl-2 messenger RNA in human breast cancer cells. Cancer Res. 2008;68(7):2358-65.
76. Dollins CM, Nair S, Boczkowski D, Lee J, Layzer JM, Gilboa E, et al. Assembling OX40 aptamers on a molecular scaffold to create a receptor-activating aptamer. Chem Biol. 2008;15(7):675-82.
77. Pastor F, Kolonias D, McNamara II JO, Gilboa E. Targeting 4-1BB costimulation to disseminated tumor lesions with bi-specific oligonucleotide aptamers. Mol Ther. 2011;19(10):1878-86.
78. Prodeus A, Abdul-Wahid A, Fischer NW, Huang EH, Cydzik M, Gariépy J. Targeting the PD-1/PD-L1 immune evasion axis with DNA aptamers as a novel therapeutic strategy for the treatment of disseminated cancers. Mol Ther. 2015;4.
79. Parashar A, Maheshwari V, Shrivastava A, editors. Enhanced n-wheeler post accidental rescue system connecting to the correct person at the correct time. 2016 IEEE Region 10 Humanitarian Technology Conference (R10-HTC); 2016: IEEE.
80. Kim M, Kim D-M, Kim K-S, Jung W, Kim D-E. Applications of cancer cell-specific aptamers in targeted delivery of anticancer therapeutic agents. Molecules. 2018;23(4):830.
81. Song Y, Zhu Z, An Y, Zhang W, Zhang H, Liu D, et al. Selection of DNA aptamers against epithelial cell adhesion molecule for cancer cell imaging and circulating tumor cell capture. Anal Chem. 2013;85(8):4141-9.
82. Shigdar S, Lin J, Yu Y, Pastuovic M, Wei M, Duan W. RNA aptamer against a cancer stem cell marker epithelial cell adhesion molecule. Cancer Sci. 2011;102(5):991-8.
83. Eilers A, Witt S, Walter J. Aptamer-Modified Nanoparticles in Medical Applications. Adv Biochem Eng Biotechnol. 2020;174:161-93. https://doi.org/10.1007/10_2020_124.
84. Li L, Xiang D, Shigdar S, Yang W, Li Q, Lin J, et al. Epithelial cell adhesion molecule aptamer functionalized PLGA-lecithin-curcumin-PEG nanoparticles for targeted drug delivery to human colorectal adenocarcinoma cells. International Journal of Nanomedicine. 2014:1083-96.
85. Shen C, Zeng K, Luo J, Li X, Yang M, Rasooly A. Self-assembled DNA generated electric current biosensor for HER2 analysis. Anal Chem. 2017;89(19):10264-9.
86. Nimjee SM, White RR, Becker RC, Sullenger BA. Aptamers as Therapeutics. Annu Rev Pharmacol Toxicol. 2017;57:61-79. https://doi.org/10.1146/annurev-pharmtox-010716-104558.

87. Ferreira C, Matthews C, Missailidis S. DNA aptamers that bind to MUC1 tumour marker: design and characterization of MUC1-binding single-stranded DNA aptamers. Tumor biology. 2006;27(6):289-301.
88. Vázquez-González M, Willner I. Aptamer-Functionalized Hybrid Nanostructures for Sensing, Drug Delivery, Catalysis and Mechanical Applications. Int J Mol Sci. 2021;22(4). https://doi.org/10.3390/ijms22041803.
89. Zhang J-J, Cheng F-F, Zheng T-T, Zhu J-J. Versatile aptasensor for electrochemical quantification of cell surface glycan and naked-eye tracking glycolytic inhibition in living cells. Biosensors and Bioelectronics. 2017;89:937-45.
90. Green LS, Jellinek D, Jenison R, Östman A, Heldin C-H, Janjic N. Inhibitory DNA ligands to platelet-derived growth factor B-chain. Biochemistry. 1996;35(45):14413-24.
91. Willis MC, Collins B, Zhang T, Green LS, Sebesta DP, Bell C, et al. Liposome-anchored vascular endothelial growth factor aptamers. Bioconjugate chemistry. 1998;9(5):573-82.
92. Ming X, Qiu S, Liu X, Li S, Wang Y, Zhu M, et al. Prognostic role of tenascin-c for cancer outcome: a meta-analysis. Technol Cancer Res Treat. 2019;18:1533033818821106.
93. Kim GR, Choi JM. Current Understanding of Cytotoxic T Lymphocyte Antigen-4 (CTLA-4) Signaling in T-Cell Biology and Disease Therapy. Mol Cells. 2022;45(8):513-21. https://doi.org/10.14348/molcells.2022.2056.
94. Trinchera M, Aronica A, Dall'Olio F. Selectin Ligands Sialyl-Lewis a and Sialyl-Lewis x in Gastrointestinal Cancers. Biology. 2017;6(1):16.
95. Ding F, Gao Y, He X. Recent progresses in biomedical applications of aptamer-functionalized systems. Bioorg Med Chem Lett. 2017;27(18):4256-69. https://doi.org/10.1016/j.bmcl.2017.03.032.
96. Heydari-Bafrooei E, Shamszadeh NS. Electrochemical bioassay development for ultrasensitive aptasensing of prostate specific antigen. Biosensors and Bioelectronics. 2017;91:284-92.
97. Nonaka Y, Yoshida W, Abe K, Ferri S, Schulze H, Bachmann TT, et al. Affinity improvement of a VEGF aptamer by in silico maturation for a sensitive VEGF-detection system. Anal Chem. 2013;85(2):1132-7.
98. Woldekidan HB, Woldesemayat AA, Adam G, Tafesse M, Thimiri Govinda Raj DB. Aptamer-Based Tumor-Targeted Diagnosis and Drug Delivery. Adv Exp Med Biol. 2023;1409:173-92. https://doi.org/10.1007/5584_2022_732.
99. Yan AC, Levy M. Aptamer-Mediated Delivery and Cell-Targeting Aptamers: Room for Improvement. Nucleic Acid Ther. 2018;28(3):194-9. https://doi.org/10.1089/nat.2018.0732.