Nucleic acid delivery: Are you developing what Big Pharma seeks?

By Dr. Daniel Sieiro & Richard A. Brown
September 2021

PLEXUS VENTURES
Business Builders in Healthcare

Abstract: *Gene therapies and other nucleic acid-based therapeutics, recently boosted by COVID-19 mRNA vaccines' resounding success, are on an upwards growth trajectory. However, these molecules' properties mean special delivery mechanisms are required for them to efficiently provide a therapeutic effect. In recent years, technological solutions by innovative Biotechs have produced a wide array of delivery vectors. Big Pharma companies are increasingly investing in nucleic acid therapeutics and are keen to partner with Biotechs to achieve targeted delivery; they expect the number of deals to significantly increase in the coming years. For a smaller Biotech looking to partner with Big Pharma, it is imperative to make the most out of limited resources in getting to this deal-making point, yet we find that Biotechs are often lost in terms of what Big Pharma expects to see. Here, we present a brief insight into what Big Pharma executives and decision makers have shared with us, in terms of what they look for when selecting a Biotech with vector technologies for the delivery of nucleic acid therapeutics.*

Objective

The objective in writing this paper is to disseminate our knowledge and perspectives on the marketplace for non-viral delivery of nucleic acids, gained from our work on behalf of Biotech producers and prospective Big Pharma buyers of these technologies. When the Biotech's technology is shown to address the wants and needs of Big Pharma, a deal typically results. Because Big Pharma has many choices when selecting a delivery technology partner, the Biotech must consider Big Pharma's current and future needs when selecting a development target.

The audience for this paper is not the development scientist working in the field, as there are excellent in-depth reviews into the structure and nature of the non-viral particles that can be found elsewhere[1–3]. Instead, our aim is to provide to the Biotech's strategic leader a behind-the-curtain perspective into the thinking of Big Pharma scientific management and thus their search & evaluation teams when it comes to appraising these technologies.

- What does Big Pharma expect to see from a prospective partner?
- What do they wish to have the Biotech incorporate into their programs?
- What advice would they give Biotechs looking to partner with them on a delivery technology platform?

Through our business development consulting experience "Business Builders in Healthcare", we continue to offer a wide range of strategic insights gained from past projects and honed by leveraging our industry sources, including from Top 10 multinational pharmaceutical and biotechnology companies. Although the focus of this paper is on non-viral vectors for nucleic acid delivery, many of the insights noted herein can also apply to other technologies where a Biotech is seeking to address problems in therapeutic delivery.

The Science: Nucleic Acid Therapies

The notion that nucleic acids could be effective therapeutics against a wide range of diseases has gone from a promising theory to an exciting reality. If further proof were needed of the potential of these therapeutics, the two recently approved COVID-19 mRNA vaccines by Pfizer/BioNTech and Moderna, which have shown notable effectiveness in disease prevention along with exceptional safety, have rightly sent this field's development into warp speed. At present, there are a total of 10 oligonucleotide products in the U.S. market and more than 100 clinical trials recruiting patients, with many more announced in the pipeline[1,4].

In contrast to conventional drugs, which generally target proteins, nucleic acid-

based therapeutics exploit the cell's genetic machinery by modulating gene expression. Introducing exogenous nucleic acids into cells to counteract defective genes, produce antigen proteins, or modify genetic sequences, among myriad possibilities, is an effective way to achieve specific and possibly curative therapeutic effects in inherited and acquired disorders.

However, nucleic acids as therapeutics present a challenging proposition. These molecules are particularly susceptible to degradation by enzymes, can contribute to immune reactions and have unfavorable physicochemical properties that prevent straightforward delivery into cells[3]. Thus, the effectiveness of these treatments critically depends on technologies designed to protect nucleic acids from degradation, ensuring their stability whilst in circulation, enabling distribution to the target tissue, and finally delivering effectively inside the cells[3]. Therefore, a vital element in the development of such drugs has been figuring out safe, effective and scalable delivery mechanisms.

Viral vectors have been an obvious candidate for delivery vehicles, as viruses have naturally evolved to effectively deliver nucleic acid material into cells, including specific cell types. However, viral delivery vehicles have encountered numerous roadblocks, from serious immunogenicity and genotoxicity to cargo size constraints, among others. Solutions to these problems are being investigated[5], but alternatives, namely non-viral delivery systems, are coming to the fore.

The development of an effective and safe non-viral gene vector is a daunting proposition, as it requires comprehensive knowledge of physicochemical and biological properties of the genetic material and carrier, the physiology of the target cells, and a mechanistic understanding of vector-induced transfection at the molecular level[4]. For this reason, pharma companies of all sizes and capabilities have been keen and willing to partner up with smaller biotechs that may bring new technologies and a fresh take at solving this intriguing puzzle. They recognize that there is a lot of current awareness and communications about the delivery of nucleic acids in the public press, especially with the advent of RNA vaccines in the wake of the COVID-19 crisis. To Big Pharma, non-viral delivery platforms are an attractive and crowded area, one where they expect to see an important increase in deals over the next three to five years.

Solving the Delivery Problem

Most currently approved or late-stage nucleic acid therapeutics rely on three main non-viral platform delivery technologies: 1) chemically modified anti-sense oligonucleotides (ASOs), 2) N-acetylgalactosamine (GalNAc) ligand-modified RNA conjugates and 3) nanocarriers, including lipid nanoparticles (LNPs) and others[3]. Compared with viruses, non-viral vectors are low in cytotoxicity, immunogenicity, and mutagenesis[4]. The challenge in this area has been to produce delivery mechanisms that protect nucleic acids from degradation, while ensuring the right cell types are targeted, something viruses do naturally.

One approach has been to chemically modify nucleic acids in the absence of a protective delivery vector, leaving them "naked" so to speak, while ensuring they are stable and resistant to nucleases and other degrading enzymes[6]. Extensive progress has been made in nucleoside chemistry: second-generation modifications include 2'-O-methyl (2'-OMe), 2'-O-methoxyethyl (2'-MOE) and others[7]. These 2' substituents influence oligonucleotide molecular conformation, resulting in improved RNA target binding affinity and mostly

increased nuclease resistance[7]. Chemical modification has been applied to various nucleic acid configurations, most commonly ASOs (anti-sense oligonucleotides), but as well to siRNAs and others. ASOs and siRNAs share important similarities as drug candidates. Both platforms are intended to modulate gene expression. Both are nucleic acids and contain an antisense strand intended to recognize a target mRNA. They also have important differences. ASOs have one strand while siRNAs have two[8].

Another currently used approach to non-viral delivery has been the use of GalNAc conjugates. GalNAc conjugation represents an efficient way of increasing siRNA target organ accumulation and of facilitating their cellular uptake. GalNAc is ideal for hepatic siRNA delivery; it targets the ASGPR receptor, predominantly expressed on hepatocytes, thus providing access to a defined cell type within the liver[9]. Furthermore, its high internalization and recycling rate allow continuous uptake of siRNA molecules, thereby increasing target cell concentration. As such, GalNAc conjugation is now one of the leading strategies for delivering oligonucleotides currently in development. However, its strong liver tropism has limited its applications, given that other tissues cannot be targeted using this system.

Nanocarriers are defined as nanoscale formulations or devices, which are able to carry medicinal drugs to targeted sites by controlling and/or targeting drug delivery. Nanocarrier compositions can vary widely, and include polymers, lipids, inorganic particles, or combinations of different types. Of these, lipid nanoparticles are arguably the most common, including as COVID-19 mRNA vaccine formulations.

LNPs typically encompass four components: ionizable cationic lipids, phospholipids, cholesterol and polyethylene glycol (PEG)-lipids. COVID-19 vaccines were no different, both containing an ionizable lipid positively charged at low pH (enabling RNA complexation) and neutral at physiological pH (reducing the potential toxic effects and facilitating payload release). They also contain a PEGylated lipid to reduce antibody association by serum proteins and clearance by phagocytes thus conferring longer systemic circulation[10].

Nanocarriers, including LNPs, represent a non-viral delivery that in contrast to chemical modifications and conjugates, provides a protected compartment, sequestered from serum nuclease activity and immune components, and a drug-biodistribution profile dictated by the carrier, not the molecule itself. These characteristics have made nanocarriers an attractive proposition for targeting non-hepatic tissues safely and precisely. Presently, nanoparticle-based mRNA vaccines are being developed against a variety of infectious diseases, including Zika virus, cytomegalovirus, tuberculosis, and influenza[10], as well as cancer immunotherapies against melanoma, ovarian cancer, breast cancer and other solid tumors[11].

Big Pharma's Expectations in Partnering

An issue we commonly encounter is a disconnect between what Biotechs see as sufficient proof of their technology's capabilities, and what Big Pharma expects when presented with a new delivery platform. One must remember that Big Pharma fields an enormous number of propositions on an almost daily basis, and often those that attract their full attention are not the ones with the wildest claims, but instead those with the most solid data. That means hitting some targets that pharma considers necessary for conversations to advance.

When considering delivery systems for nucleic acid therapeutics, the primary question

Target tissue delivery and expression data are no longer enough for Big Pharma, cellular-level data is now widely expected.

executives spoke most about was: "what are the tissues being targeted, and how solid is the proof?" As is well known, a major obstacle preventing widespread usage of oligonucleotide therapeutics is the difficulty in achieving efficient delivery to target organs and tissues other than the liver[1]. With the success of GalNAc and others, liver-targeting technologies are not seen as distinguishing as they may have once been; there are already tried and tested mechanisms to choose from so the need for something new is less pressing.

For technologies aiming to target extra-hepatic tissues, executives reveal that an approach demonstrating tissue expression at or below 3x over baseline liver expression levels is now commonplace and is not likely to stand out. Instead, a true differentiator would have to be 5x higher expression in the target tissue when compared to the liver ("Your technology would then be hot!"). If the vectors are likely to go to non-target tissues due to, for instance, systemic administration (i.e., standard distribution to other tissues outside the target), then one needs to confirm that sequences are not expressed there by conducting biopsies.

Proven targeted tissue delivery and expression are no longer enough for Big Pharma to be interested: cellular-level data is now widely expected. Companies want to see that the right cell types within the target tissue are being hit, that the cargo is being safely delivered within those specific cells, that endosomal escape is not an issue, etc. Executives acknowledge that cellular data is not yet commonplace for companies approaching them, but they reveal that for any discussions to advance, they expect nothing less. While this creates a higher barrier to entry, it opens up certain possibilities. New strategies in liver delivery are of interest in this case, since GalNAc targets almost exclusively hepatocytes[12], while there is still a plethora of hepatic cell types linked to various other diseases.

As would be expected, any non-viral vector presented will need to show proof-of-concept data that verifies efficacy. In particular, Big Pharma wants to see study comparisons with other direct competitors to show any advantages. They may even require the Biotech to conduct a test with housekeeping genes or a gene of Big Pharma's choosing to verify the feasibility of the vector delivery. This will most likely be an expense borne by the Biotech itself and not in partnership with Big Pharma. Any proposal will also need to provide strong proof-of-concept (PoC) safety data showing a lack of toxicity due to an immunogenic response. While non-viral gene vectors possess lower immunotoxicity than viral vectors, it should not be overlooked that

they can trigger immune reactions[13]. Thus, executives see a history of toxicity of related technologies as a major worry. Finally, for both efficacy and safety data, including PoC and bio-distribution, executives conveyed they would prefer to see data in non-human primates, as mice results are not so impressive given their considerably smaller body size.

For nanoparticles in particular, size is a key factor that affects all steps in the delivery process. Generally, a larger size is favored for long circulation time in the blood, but a relatively smaller vector can more efficiently permeate cells[4]. Particle size can also affect the cellular uptake mechanism and rate[14]. Therefore, cargo is an important matter, especially with larger payloads such as gene-editing. For Big Pharma decision makers, size limitations for a payload are important, as they prefer the ability to deliver large cargos as needed, even if their current programs do not require it. Long-term flexibility of a delivery platform is highly desired.

For a smaller Biotech, commercial product development, especially relating to a vector's physical and chemical stability during formulation preparation and product storage, is typically not a priority at early stages of development[4]. However, for Big Pharma, commercialization is always on their mind. Thus, cost of goods and CM&C (Chemistry, Manufacturing and Controls) will be important. Providing data on these matters is not strictly required but setting up technologies for future scalability will certainly give a competitive advantage. For example, if a nanocarrier can be highly tissue and cell specific, then it can be dosed at lower amounts, saving in the volume of the product used and lowering cost of goods. Likewise, the burden on CM&C to manage multiple vectors (quality, stability) limits the feasibility of licensing them on a compound-by-compound basis. Thus, a company providing a platform technology that covers various compounds will take precedence over others, from a commercial standpoint.

Big Pharma's Wish-list

The search for a Big Pharma partner interested in a particular delivery technology for their own internal programs is somewhat of a blind matchmaking exercise. Big Pharma's openness to considering something novel will likely be based on whether their internal programs are hitting their targets or not. Unfortunately, most times these are confidential conversations, so knowing whether companies are in need of new ideas may be close to impossible. However, there are certain criteria that Big Pharma executives have acknowledged will get their attention whether they have immediate needs or not.

While speaking with Big Pharma decision makers, there were certain target tissues that spiked their interest. We often heard enthusiasm for any vectors that specifically target the heart, particularly myocardial tissue; other muscle tissue types were also mentioned. Specific delivery to areas such as pancreatic beta cells was high on their list. As expected, there was often mention of non-viral vectors that can efficiently and demonstrably cross the blood-brain barrier. Likewise, the ability to deliver to hematopoietic stem cells in bone marrow is seen as a "game changer", as it would replace *ex vivo* therapy for *in vivo* methods. For some companies, administration of non-viral vectors through sub-cutaneous delivery would represent a breakthrough, with one executive calling it a "Holy Grail". AstraZeneca's recent announcement of subcutaneous administration of mRNA using anti-inflammatory steroid precursors[15] reiterates that interest.

Big Pharma is constantly looking for delivery

Non-viral vectors that can efficiently and demonstrably cross the blood-brain barrier or deliver to hematopoietic stem cells in bone marrow are seen as game changers.

technologies that will allow for a more efficient and tightly controlled pipeline, with less variability among programs. To them, the question when encountering novel systems is: "does this vector have to be switched out every time you change molecule type?" Thus, a Biotech that can show data on their technology with several different cargos, behaving consistently, will most likely have an advantage. Of course, companies know that delivery systems are rarely a one-size fits all approach. Nanoparticles' cocktail composition must be tailored to each situation for example. However, if the technology's owner can show a mastery of their formula, conveying a sense of adaptability to whatever Big Pharma may want to carry, this will instill a sense of confidence in concluding a deal.

Big Pharma's Essential Advice

Aside from sharing requirements and wish lists, Big Pharma executives candidly disclosed advice they would give Biotech companies thinking of approaching them with a platform technology. Importantly, Big Pharma wants to deal with a company that has a delivery "platform". As mentioned above, they defined their preferred platform as one that can deliver more than one type of nucleic acid to a targeted organ or cell type by slight modifications of the delivery system, not a variety of different vectors that can deliver cargoes to multiple tissues. The implication here is that, although the Biotech may identify methods of achieving specificity to multiple organs, to a single licensee this may not be appealing, since they may be focusing on a single or a few organs at most.

Executives expect the delivery vector company to be highly specialized in a certain type of delivery (better than the competition), such as being experts in delivery to the heart for example, not to all tissues. They state that too many companies try to do too many things and have a lack of focus. Overall, their advice to small Biotechs is to focus on what works best with their technology and stick with that; if their vector naturally works best on heart tissue for example, build everything around that. In their perspective, a vector that can deliver many different cargos to a single tissue with high specificity and safety will be more desirable than a vector that can deliver a single cargo to several tissues with average efficiency. As such, when contacting pharmaceutical companies, a focused offering will generate more interest than a broad capability, at least initially. Conducting an analysis of the Big Pharma's pipeline and evidence of research focus should allow the Biotech to anticipate, to a large degree, the type of challenges they may be encountering. Each company will

> *Big Pharma prefers to deal with a company that is open to discuss both the advantages and limitations of their technology. Openness creates trust.*

have specific needs, which may consist of an individual organ or cell-type. It is then important to explain from the outset how the team (people + technology) has shown the capability to solve the prospective partner's needs.

When approaching a potential partner, there are other strategies that will help smooth the process and increase the chance of an initial positive response. Big Pharma decision makers prefer to deal with a company that is open to discuss both the advantages and limitations of their technology, and to disclose enough about the "secret sauce" in a prospective way so that Big Pharma does not have to extract the specific details little by little through continued questioning. Openness creates trust.

As such, during initial discussions, they prefer that the company describe their technology and how and why it works and why it is better than the competition, with hard data and not flashy slides. Similarly, the Biotech should be prepared to disclose their methodology for optimizing the delivery vectors (under CDA of course). Since Big Pharma companies will select their partners based on the likelihood of being successful on their internal assets, any experience in achieving a successful result will build confidence in being able to overcome future development challenges. It is important to show that the Biotech has optimized the delivery method, and that it can logically translate to a new formulation specific to a compound of importance to the Big Pharma.

This, however, does not mean glossing over any encountered problems and showing only the successes. Big Pharma will appreciate the potential partner to show the process it took to get to the current product, not just the end result. For them, seeing this process, and how issues were resolved, shows a Biotech's ability to critically think their way to a solution, so that they, if entering into a partnership, will be able to resolve any issues that might arise when dealing with the Big Pharma's molecules and/or cell targets.

Conclusion

The good news is that with nucleic acid therapeutics becoming more prevalent, the demand for effective, efficient and safe delivery platforms will continue to be strong. Big Pharma companies will be out searching for those innovative technologies that will enable their compounds to reach their therapeutic potential, thus the number of deals is expected to increase, with the value paid based on how uniquely the technology fits the application.

It is incumbent upon the Biotechs developing these technologies to anticipate likely

applications and prepare the evidence that demonstrates the desired attributes. Here we have provided a glimpse of what a sample of Big Pharma expects to be shown, but this is by no means a global picture. Arranging recurring conversations with Big Pharma decision influencers throughout the development of the technology is the best way to understand the needs and thereby guide the development program. Executed effectively, the Biotech should be in an excellent position to pursue partnerships with Big Pharma, armed with persuasive data supporting the correct fit to a range of Big Pharma's delivery requirements.

Dr. Daniel Sieiro is Manager for Innovation based at the Boston and Paris offices. Richard A. Brown is Managing Partner based at the Indianapolis and Tokyo offices.

About Plexus Ventures

Plexus Ventures LLC is a global business development organization with a unique set of capabilities and an extensive international network. The team is comprised of 12 senior business development executives who have previously served in companies such as GlaxoSmithKline, Johnson & Johnson, Eli Lilly, Merck, Novartis, Menarini and Recordati, as well as Harvard Medical School in research capacities. Plexus' team includes three PhDs, one MD and an array of commercial talent. Plexus Ventures' senior management team (Bob Moran, Michael O'Sullivan, Richard Brown and Gabriele Tundo) are strategically located in Philadelphia, London, Indianapolis/Tokyo and Milan.

Plexus Ventures has successfully served its pharmaceutical, consumer healthcare and drug delivery clients for 30 years. During this time, they have conducted all types of business development projects, specializing in providing strategic advice as part of every client assignment in the areas of in-licensing, out-licensing, acquisitions and divestitures. Their confidential process and contacts follow a strict methodology in order to create a sense of competition and urgency amongst potential partners. Ultimately, Plexus' goal is to perform transactions for clients in a timely manner obtaining the highest value possible.

Plexus has conducted successful transactions for companies both large and small, having worked successfully for AstraZeneca, Eisai, GSK, Johnson & Johnson, Kyowa Kirin, Mabion, Novartis, Glenmark, to name a few. In the areas of nucleic acid therapeutics and delivery, Plexus has most recently supported Crystal Therapeutics on a polymer nanoparticle technology platform, two projects for Nitto Denko on their lipid nanoparticle technology platform, as well as on behalf of ProQR for the out-licensing of their ASO QRX-704 for the treatment of Huntington Disease.

Please direct any questions relating to this article or other information requests to the corresponding author, listed below.

Daniel Sieiro
Manager, Innovation
Boston and Paris Offices
Mob US: +1 617 331 4794
Mob FR: +33 (0) 7 66 77 3538
Email: daniel_sieiro@plexusventures.com

PLEXUS VENTURES
Business Builders in Healthcare

References

1. Roberts, T. C., Langer, R. & Wood, M. J. A. Advances in oligonucleotide drug delivery. *Nat Rev Drug Discov* 19, 673–694 (2020).
2. Hammond, S. M. et al. Delivery of oligonucleotide-based therapeutics: challenges and opportunities. *Embo Mol Med* 13, e13243 (2021).
3. Kulkarni, J. A. et al. The current landscape of nucleic acid therapeutics. *Nat Nanotechnol* 16, 630–643 (2021).
4. Zu, H. & Gao, D. Non-viral Vectors in Gene Therapy: Recent Development, Challenges, and Prospects. *Aaps J* 23, 78 (2021).
5. Capra, E., Godfrey, A., Loche, A. & Smith, and J. Gene therapy innovation: Unlocking the promise of viral vectors. *McKinsey & Company* (2021).
6. Springer, A. D. & Dowdy, S. F. GalNAc-siRNA Conjugates: Leading the Way for Delivery of RNAi Therapeutics. *Nucleic Acid Ther* 28, 109–118 (2018).
7. Manoharan, M. 2'-Carbohydrate modifications in antisense oligonucleotide therapy: importance of conformation, configuration and conjugation. *Biochimica Et Biophysica Acta Bba - Gene Struct Expr* 1489, 117–130 (1999).
8. Watts, J. K. & Corey, D. R. Silencing disease genes in the laboratory and the clinic. J Pathology 226, 365–379 (2012).
9. Nair, J. K. et al. Multivalent N Acetylgalactosamine-Conjugated siRNA Localizes in Hepatocytes and Elicits Robust RNAi-Mediated Gene Silencing. *J Am Chem Soc* 136, 16958–16961 (2014).
10. Tenchov, R., Bird, R., Curtze, A. E. & Zhou, Q. Lipid Nanoparticles—From Liposomes to mRNA Vaccine Delivery, a Landscape of Research Diversity and Advancement. *Acs Nano* (2021) doi:10.1021/acsnano.1c04996.
11. Gómez-Aguado, I. et al. Nanomedicines to Deliver mRNA: State of the Art and Future Perspectives. *Nanomaterials-basel* 10, 364 (2020).
12. Prakash, T. P. et al. Targeted delivery of antisense oligonucleotides to hepatocytes using triantennary N-acetyl galactosamine improves potency 10-fold in mice. *Nucleic Acids Res* 42, 8796–8807 (2014).
13. Jones, C. H., Chen, C.-K., Ravikrishnan, A., Rane, S. & Pfeifer, B. A. Overcoming Nonviral Gene Delivery Barriers: Perspective and Future. *Mol Pharmaceut* 10, 4082–4098 (2013).
14. Buck, J., Grossen, P., Cullis, P. R., Huwyler, J. & Witzigmann, D. Lipid-Based DNA Therapeutics: Hallmarks of Non-Viral Gene Delivery. *Acs Nano* 13, 3754–3782 (2019).
15. Davies, N. et al. Functionalized lipid nanoparticles for subcutaneous administration of mRNA to achieve systemic exposures of a therapeutic protein. Mol Ther - *Nucleic Acids* 24, 369–384 (2021).