Inside the Spray:

How In Vitro Tools Reveal What's Really Happening When Your Product Meets a Patient

When a nasal spray or inhaler works well in a patient, a lot of things must go right. The formulation must stay stable. The device must be atomised correctly. The resulting aerosol must travel to the right place in the airway. And all of that must happen under the unpredictable conditions of real-world use, including variable actuation, variable shaking, variable angle, a nose that might be congested, and lungs that breathe at different rates.

Performance is not a property of the formulation nor device. It emerges from the interaction between formulation, device, and how the patient uses it. That is a complex system, and complex systems can reveal their weaknesses late unless they are interrogated early.

This is what working inside the spray means: building development around the mechanisms that drive performance, not just the measurements that confirm a specification has been met.

Measurement and Understanding Are Not the Same Thing

The orally inhaled and nasal drug product (OINDP) field knows how to generate data. Spray pattern, plume geometry, aerodynamic particle size distribution, droplet size distribution, and delivered dose uniformity are described in general pharmacopeial chapters and supported by regulatory guidance, although the chapters themselves do not prescribe detailed step-by-step methods in the way some other tests do.[1–3]

The harder question is whether the data being generated is the data needed to understand performance. All data provides information, but it only becomes useful when it captures the variables that matter for how the product behaves in use.

A product can pass every required test yet still fail to perform as intended in a real patient. The gap between passing a test and working clinically is where programmes get into trouble, often quietly, and often late.[4]

The scale of that gap is quantifiable. Orally inhaled drugs carry a clinical attrition rate of approximately 70% – seven out of ten candidates that enter trials do not reach the market.[5] The cumulative probability of a respiratory drug achieving approval is just 3%, compared to 6–14% for drugs in other therapeutic areas.[6] These are products that have already passed preclinical and *in vitro* testing. The failure is not happening before the data is generated. It is happening after.

What Drives Performance

To work within the spray, you begin with a fundamental question: What physical and chemical mechanisms govern how this product performs?

Rather than simply asking whether the spray pattern meets established acceptance criteria, ask instead what happens during atomisation, how the formulation breaks apart, what droplets are formed, where they travel, and what state the drug is in when it arrives at the target tissue.

For conventional small molecule nasal sprays, those questions are answerable with established tools and methods. But the OINDP pipeline is changing. Biologic molecules, including proteins, peptides, and nucleic acid therapies, are entering inhaled and nasal development in increasing numbers. Existing drugs are also being reformulated for nasal delivery to avoid injection, improve onset, or reach the brain directly. These products are more sensitive and more complex than the formulations around which the field's standard frameworks were built.

Immunogenic and therapeutic payloads, including DNA, mRNA, peptides, and proteins, are increasingly formulated within nanoparticle carrier systems such as lipid nanoparticles (LNPs), liposomes, polymeric nanoparticles (PNPs), and virus-like particles (VLPs) for intranasal administration. Each carrier system introduces distinct physicochemical properties that influence how the formulation behaves during atomisation and what analytical approaches are required to characterise it.

For a biologic payload, the spray event itself can be a stability challenge. Shear forces during atomization can denature proteins and disrupt lipid-based systems under certain conditions.[7] Stability data on the bulk formulation does not describe what comes out of the device. Whether the molecule survives the spray intact, and how that depends on device design and actuation conditions, is a question that needs to be asked deliberately and answered with the right tools.

Deposition and Delivery Are Not the Same Thing

One of the most consequential assumptions in nasal drug development is treating what leaves the device as equivalent to where it ends up. Delivery describes what comes out. Deposition describes where it goes. The difference between them is often the primary determinant of whether the product works.

In nasal delivery, deposition depends on droplet size, velocity, spray angle, insertion depth, and airway geometry, as well as the anatomy of the nasal cavity itself, which varies

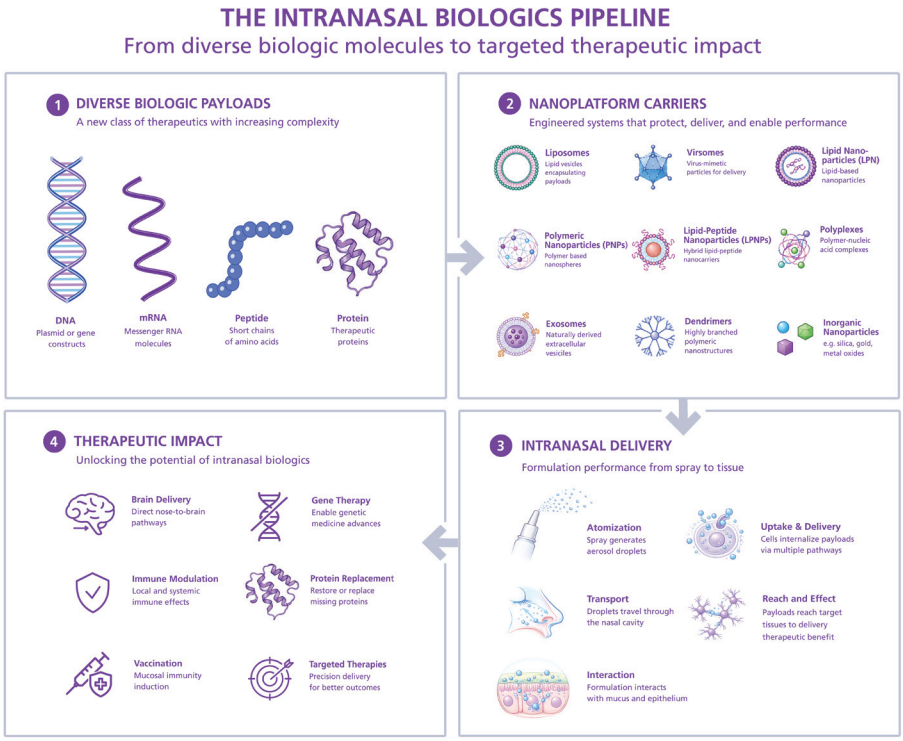


Figure 1: The intranasal Biologics Pipeline: From Biologic payload selection and nanoplatform engineering to nasal delivery, tissue interaction, and therapeutic impact.

significantly between individuals and with mucosal condition.[6,7]

One of the most persistent challenges in nasal drug delivery is anterior deposition. When a dose deposits in the nasal vestibule or the front of the nasal cavity, it is likely to drip out before it can be absorbed, producing no therapeutic effect. Getting the dose past the vestibule is the first requirement of effective nasal delivery, and it is not guaranteed by standard spray configurations.

Published nasal cast deposition studies by De Bank *et al.* demonstrated that administration technique can significantly influence regional nasal deposition, with anterior deposition observed across multiple actuation angles under many test conditions condition.[8]

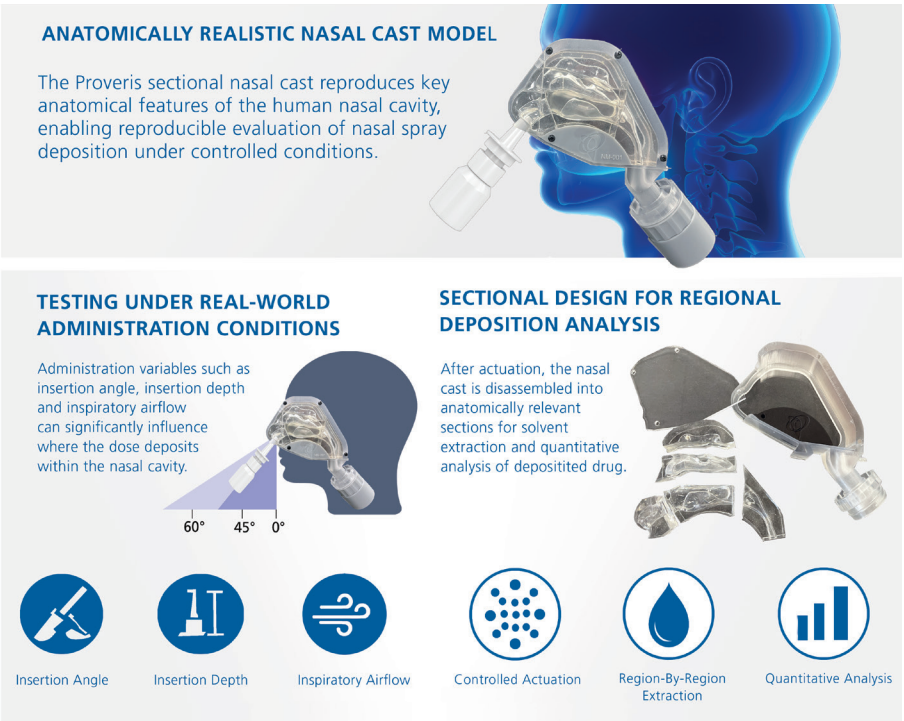


Figure 2. Anatomically realistic sectional nasal cast used to evaluate the impact of administration variables including insertion angle, airflow, and regional deposition analysis

The challenge becomes significantly greater when targeted deposition is required. For products where the therapeutic outcome depends not just on clearing the vestibule, but on the dose arriving in a specific region of the nasal cavity – like the olfactory for nose-to-brain delivery or the lymphatic associated tissue in the back of the nasal cavity – inferring where the dose goes from traditional *in vitro* tests alone is not sufficient. Testing directly in an anatomically realistic nasal cast model is the only way to know early on in development.[9,10]

The Proveris sectional nasal cast allows the model to be tested under controlled administration conditions and then disassembled into anatomically relevant regions for extraction and quantitative analysis.

The Patient Is Part of the System

Human use is complex. Patients actuate devices with different forces and velocities. They breathe at different flow rates depending on age, disease state, and effort. They insert devices at different angles. None of this is controllable in the clinic but it can be studied, characterised, and incorporated into laboratory testing in ways that make *in vitro* results meaningfully more predictive of real-world performance.[11]

For nasal sprays, actuation velocity is one of the most consequential variables in the system. It directly governs atomisation, what droplet sizes are produced, and ultimately where the dose deposits. The figure below shows the range of actuation velocities measured across a real patient population. This is not a tight cluster around a single value. It is a distribution, and a product will encounter all of it.

That variability has visible consequences for spray behaviour. Atomisation characteristics change substantially across the range of velocities a product encounters in real patient use. The plume geometry images below show the same product actuated at low and high velocity.

This variability is not something to eliminate. It is part of the product's operating environment, and understanding it is part of understanding performance. Proveris Vereo® automated actuators are designed around this reality. They can replicate the range of force, velocity, and profiles that represent real patient populations.

Closing the Loop: From *In Vitro* to *In Vivo*

Clinical studies are the ultimate test of an

OINDP product, but they are expensive, slow, and offer limited mechanistic insight into why a product performs the way it does. The question for development is how to build confidence in clinical outcome before you get there.

For inhaled products, the variables that govern deposition, particle size, flow rate, and device resistance are measurable *in vitro* when the right conditions are applied. For nasal products, the same is true: actuation velocity, droplet characteristics, and anatomical deposition patterns can all be studied in the laboratory when testing is designed around human-realistic conditions rather than idealised ones. Proveris Laboratories offers human-realistic deposition studies using nasal cast models and INVIDA®, for orally inhaled drug products. This gives development teams direct, anatomically realistic deposition data earlier in the programme before formulation direction and device specifications are locked. The connection between *in vitro* results and expected *in vivo* behaviour is traceable, not assumed.

Building that connection early is not a late-stage validation exercise. It is a risk-reduction strategy. Programmes that understand deposition behaviour before committing to formulation direction and device specifications carry less uncertainty into clinical development and encounter fewer surprises when they get there.

The Floor and the Ceiling

Regulatory frameworks provide the structure within which OINDP development operates. FDA and EMA guidance, pharmacopeial chapters including USP <601>, and bio-equivalence requirements define what must be demonstrated. They are the floor.

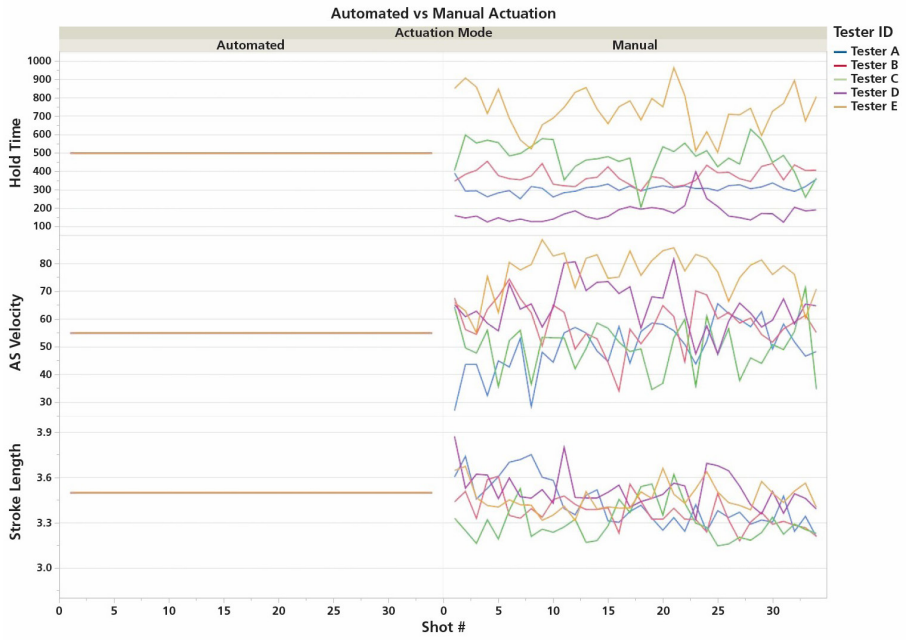


Figure 3. Consistent Control over actuation parameters with automated actuation (left) and variation observed with human actuation for pMDI devices (right)

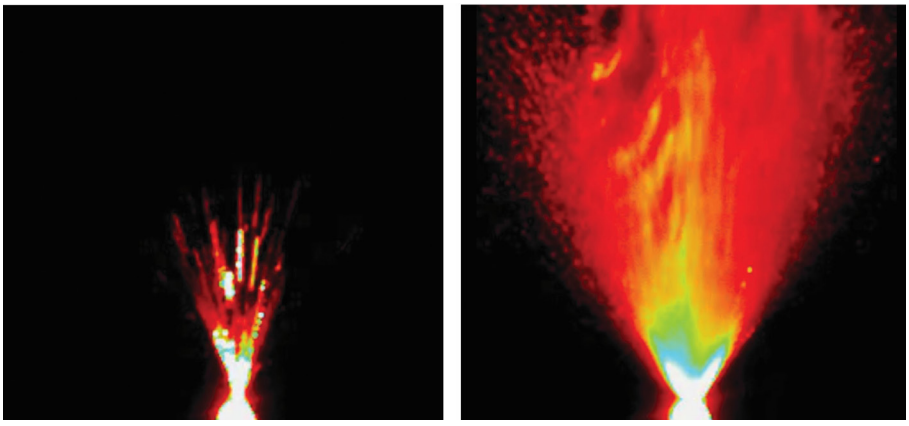


Figure 4. Plume geometry for low(left) and high (right) actuation velocities.

What they do not define is the ceiling. The programmes best positioned at submission, at scale-up, and through post-approval change management are those that treated regulatory requirements as a starting point for scientific investigation rather than its boundary. They know not just that the product meets specification, but why and what would cause it not to.

The OINDP pipeline now includes biologics, nose-to-brain therapeutics, and complex reformulations that the existing frameworks were not specifically designed for. Working inside the spray means applying those frameworks with enough rigor to know when they are sufficient and when additional understanding is required. The tools to do that work exist. Using them deliberately, from the performance drivers outward, is what responsible development in this space looks like.

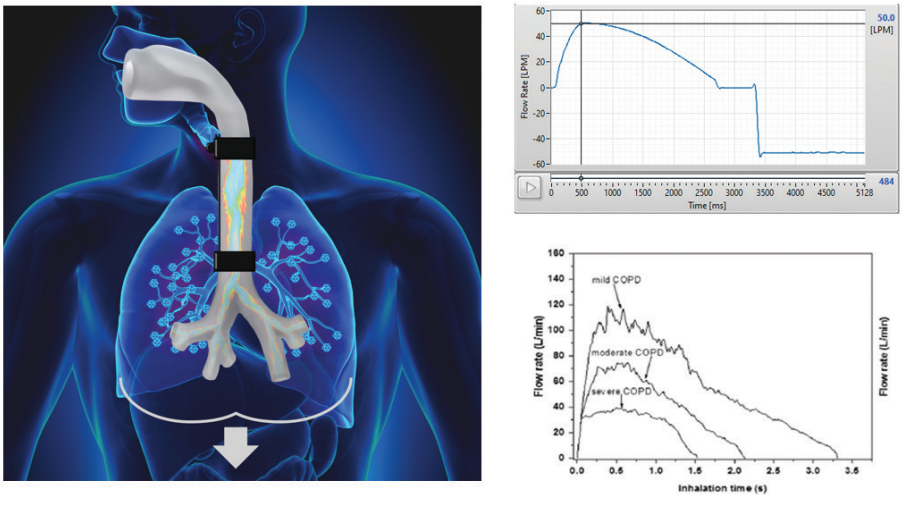
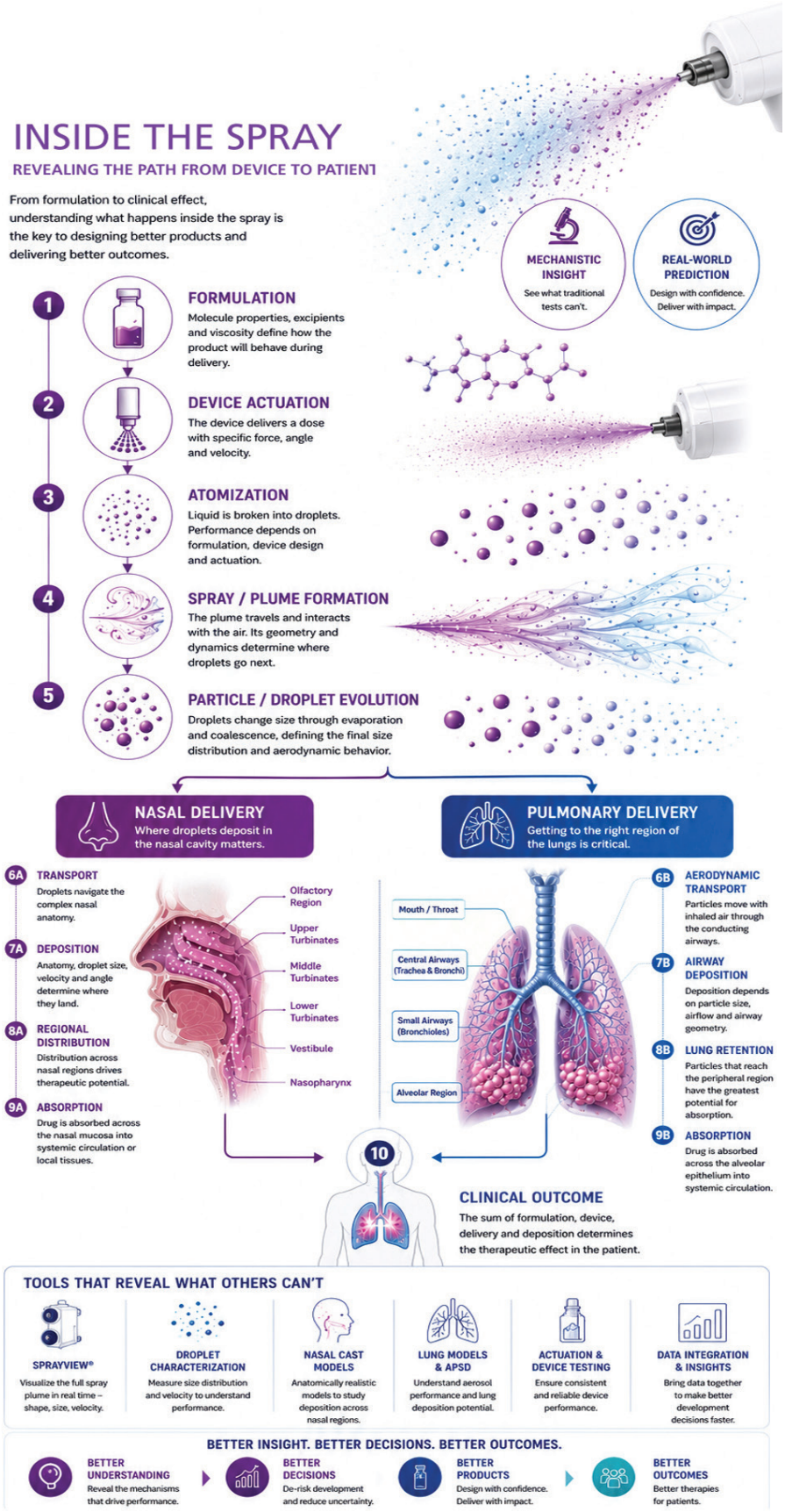


Figure 5. INVIDA In vitro respiratory model with representative breathing profiles provides indication of in vivo performance in a targeted region

REFERENCES

1. U.S. Food and Drug Administration (FDA). Guidance for Industry: Nasal Spray and Inhalation Solution, Suspension, and Spray Drug Products – Chemistry, Manufacturing, and Controls Documentation. Rockville, MD: FDA; 2002.

2. European Medicines Agency (EMA). Guideline on the Pharmaceutical Quality of Inhalation and Nasal Products (EMEA/CHMP/QWP/49313/2005 Rev. 1). London: EMA; 2006.
3. United States Pharmacopeia. General Chapter <601> Aerosols, Nasal Sprays, Metered-Dose Inhalers, and Dry Powder Inhalers. In: USP–NF. Rockville, MD: United States Pharmacopeial Convention; current edition.
4. Byron PR, Dalby RN, Hickey AJ, et al., eds. Respiratory Drug Delivery (Proceedings of the Respiratory Drug Delivery Conference Series). River Grove, IL: Davis Healthcare International; various years.
5. Priming P, Cavagnaro J, et al. Preclinical development of orally inhaled drugs (OIDs) – are animal models predictive or shall we move towards *in vitro* non-animal models? Animals (Basel). 2020;10(8):1259.
6. Prina E, Caram LMO, Soriano JB, et al. Barriers to new drug development in respiratory disease. Eur Respir J. 2015;45(5):1197–1207.
7. Maa YF, Hsu CC. Protein denaturation by combined effect of shear and air–liquid interface. Biotechnol Bioeng. 1997;54(6):503–512.
8. Maaz A, Blagbrough IS, De Bank PA. *In vitro* evaluation of nasal aerosol depositions: an insight for direct nose to brain drug delivery. Pharmaceutics. 2021;13(8):1079
9. Finlay WH. The Mechanics of Inhaled Pharmaceutical Aerosols. San Diego, CA: Academic Press; 2001.
10. Cheng YS. Aerosol deposition in the extrathoracic region. Aerosol Sci Technol. 2003;37(8):659–671.
11. Illum L. Nasal drug delivery – possibilities, problems and solutions. J Control Release. 2003;87(1–3):187–198



Joanne Mather

Joanne Mather is a scientific marketing leader with many years of experience in the analytical science space. As Senior Director of Marketing at Proveris Scientific, she focuses on translating complex scientific and regulatory challenges into practical solutions that help companies in the OINDP space accelerate development and ensure product quality. With a strong background in analytical science and a customer-centric approach, she is dedicated to supporting the industry in bringing effective and reliable aerosolized drug products to market.

Grant Thurston

Grant is responsible for overseeing the development and enhancement of Proveris product lines, ensuring the organisation continuously meets and exceeds customer expectations. Grant has a deep understanding of laboratory instrumentation and strong ability to communicate technical concepts effectively. Grant holds Bachelor and master's degrees in biomedical sciences. He is fluent in English and Spanish.