Ag Playbook

Second Edition 2026

Contents

Introduction

Welcome to the Ag Playbook!

When we released the first version back in 2024 we opened the playbook by stating:

The importance and need for innovation in agriculture cannot be overstated. But evaluating that innovation, especially in its pre-commercial stages, requires a common baseline understanding or framework for the product development process. The goal of this document is to deliver a Playbook to help startups, investors, and industry think about what stage a product is at in its development and understand and plan for the cost and effort needed to bring it to market. In doing so, this Playbook aims to aid the efforts of AgTech entrepreneurs so they can more effectively bring the next generation of ag solutions to sustainably feed the world.

Every word remains true today. Since the release of the Playbook, we have continued to face challenging market dynamics for entrepreneurs and investors alike. However, to my pleasant surprise, we have also seen a new wave of startups emerge, and the terminology used to describe their technology has begun to standardize. We have witnessed the emergence of thoughtful timelines, effective fundraising strategies, and successful product registrations.

Building on this momentum, we are excited to expand upon the original work by introducing three new sections in Ag Playbook 2.0.

- Two new introduction sections:
    - The first is a story arc about the past, present, and future of the AgTech capital markets: **The Six 'R' Eras of AgTech** from the brain of Scott Porter
    - The second a focus on the importance of **Regulatory Strategy** from Jerry Hjelle, who has helped countless Ag products make their journey through the numerous international governing bodies.
- New chapter on **Crop Protection: Biomolecules** written by three entrepreneurs who've worked deeply in the space: Thomas Laurent, Patrice Selles, and Andy Renz

With these new sections we hope to broaden the scope of topics related to AgTech product development and further round out this Playbook to move the entrepreneurs in our industry an incremental step closer to achieving our shared dream of improving the AgTech and Food ecosystem.

I would like to extend my sincere gratitude to all the volunteers who contributed to Ag Playbook 2.0. Their efforts have not been compensated financially but stem from the hope of increasing the probability of success for entrepreneurs, investors, and farmers alike. These contributors and reviewers embody the spirit of my favorite quote from the Farmer's Almanac:

"It is by our works and not by our words we would be judged; these we hope will sustain us in the humble, yet proud station we have so long held."

-PJ Amini, Editor

Methods & Review

This Playbook is the result of contributions by numerous experts who have worked deeply in the field of agriculture R&D and product commercialization. The opinions expressed here are the individual's own and do not reflect the view of their employer(s).

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The Problem[1]

Bringing a novel agricultural technology (AgTech) product to market is hard. Over the last 10 years we've seen hundreds of products enter the commercial market and into the toolkit for crop farmers. However, a large percentage of these products came from five large agricultural companies (Bayer, BASF, Syngenta, Corteva, FMC) who have spent billions in discovery, development, securing regulatory approval, manufacturing, and ultimately selling these solutions in the market. This is a high bar to set for AgTech startups. If startups plan to follow the conventional industry approach, they will need additional capital, manpower, and expertise to develop and deliver a new product. This makes startups turn to venture capital investment to fund the stages of R&D until they are ready to commercialize their innovation. The AgTech sector is not unique in its use of venture capital, however, it has yet to achieve a common understanding across innovators, funders, and established players of the time and cost it can take to support new products. Once a product is finally ready for commercial launch, AgTech startups encounter a second problem in trying to scale the solution: it becomes clear that getting onto one, ten, or a hundred million acres is very costly, unless the startup partners with other agricultural companies and retailers. Without leveraging the network and existing customer base of these partners, it can be difficult and prohibitively costly to reach the farmer customer. This is the **Reverse Field of Dreams** scenario: "If you build it, the market will not necessarily come."

In short, there are two central problems: 1) the time and cost it takes to develop agriculture products and 2) the difficulty of reaching sufficient customer numbers absent partnering with existing incumbents. Given these challenges, what are the appropriate milestones and inflection points for investors and startups? This Playbook is aims to address the first problem.

Take crop protection small molecule products as an example (not including any of the RNAi or peptide-based products). From 1995 to 2019, the cost of developing a small molecule product increased by 99% from $152m to $301m and the path to market lengthened from 8.3 to 12 years [1]. It now takes even longer to take a potential product through the process of hit finding → lead optimization → toxicity screening → greenhouse/field → regulatory trials → market development trials. Even with the acceleration of discoveries of new hits through use of artificial intelligence (A.I.) and computational platforms and tools, we still see the time it takes to get to market extending due to growing requirements from regulatory agencies. Additionally, there is the time it takes to partner with farmers, academics, trade groups, and established companies to ensure that you achieve not just efficacy but also **belief**. Belief, in this case, is defined as customer acceptance and success. Even if a product makes it through the R&D pipeline, farmer adoption is not guaranteed without industry trust. Farmers operate a high-risk, single decision-point business each growing season that can make trusting a new product difficult. Belief in a product's potential is essential in the AgTech market and is a critical factor in determining which products find success or failure.

[1] Please note that much of the problem statement revolves around plant agriculture. While many of the challenges are similar in animal agriculture, the examples used have been simplified to solutions for crops.

What's different between AgTech & Tech?

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Not all products and product pathways in agriculture follow the same process, but the example above of crop protection small molecule products demonstrates the general trend in the industry of increasing cost and time to market. Developing novel and efficacious products for agriculture is difficult. Safety and regulatory approval are paramount for these products, and unfortunately the digital startup method of building minimum viable products doesn't apply. In the regulated product market of AgTech that often requires government agency approval prior to marketing, the metaphor of "building the skateboard" and delivering it to the market then getting feedback before building the "bike," and ultimately the "car" is not viable, especially when rigorous regulatory testing needs to be conducted and reviewed to support product registrations. In the case of new pest control technologies, products follow a path where they are tested in the lab, growth chamber, and greenhouse prior to being tested in the field. Each phase of testing allows for efficacy and spectrum of control to be tested safely but delays the time to market. The volumes of material needed to test at even the early phases often require access to mini or pilot scale manufacturing capabilities. This means practically, it may be years into product development before investors receive their first actual in-field data or user feedback on results.

What's different between AgTech & Pharma?

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There are parallels between the R&D pathways of AgTech and pharmaceutical (pharma) products. Like AgTech, pharma is a highly regulated market, with the Food & Drug Administration (FDA) and its equivalent global organizations playing a paramount role in ensuring the safety of products brought to patients across the world.

However, there are **five primary differences** when comparing AgTech product development to pharmaceutical product development (taking a US centric view below):

1 Regulatory complexity

AgTech is much like Pharma in that, it develops regulated products that require the approval or consultation of government agencies. In the U.S., depending on the product type, approval by the Environmental Protection Agency (EPA), and United States Department of Agriculture (USDA) may be required in addition to state agency registrations. The U.S. Fish and Wildlife Service (FWS) also enforces national policies around product safety associated with the Endangered Species Act (ESA). Depending on product type, like Pharma, FDA approval or consultation may also be needed. But it doesn't end there. Many ag products are traded in international commerce, so national import approvals are often required in countries where the commodity will end up in addition to the local approvals required where the products are initially grown. Today there is a still variation amongst the testing protocols for different national agencies adding complexity to international scale-up.

2 Seasonality

Each growing region has a limited timeframe when crops can be successfully planted, grown, and harvested, driving the patterns of supply and demand for agricultural products. In AgTech R&D this also means there is limited time when new products can be field tested. Most geographies north or south of the 30° latitude lines have a single main growing season, with planting in the spring and harvesting in the fall. This translates to one chance a year to test a new product during the growing season in those geographies. Researchers can innovate around this limitation by running counter-season programs in northern and southern hemispheres where the seasons are staggered or by conducting research trials in geographies that have multiples seasons (e.g. Mexico). However, if a researcher discovers a new product for the North American market in October, they will likely have to wait six months before beginning a field trial. The impact of seasonality can be very challenging for many early-stage companies working on AgTech innovations.

3 Quick on efficacy, slow on safety

Initial testing for efficacy can be far quicker on the agriculture side, particularly for crop protection products due to being able to test target species such as weeds, insects, and fungi quickly. A scientist looking to develop a novel form of pest control can quickly determine if a crop protection product is affecting/inhibiting growth of the target. However, AgTech products also must prove their effect (or lack thereof) not only in the target organism, but also in a wide array of non-target organisms found in the environment as well. While Pharma products are also tested in non-human species, AgTech product safety testing requirements extend into a highly diverse species set including crop and native plants, mammals, birds, insects, and reptiles.

4 Belief through engagement

Engaging directly with the farmer who buys the product is required in AgTech. Farmers make independent and informed decisions and are not limited in their product access the way patients may be with many Pharma products due to needing insurance coverage and a doctor prescription. Farmers are highly knowledgeable of product offerings and their business needs, engage expert agronomists, and are looking for consistent data and results that support product performance. Because of the wide variability in fields, weather, and agronomic practices, engagement early on to share efficacy and safety to build belief is critical for a successful product launch. Working with groups like the United Soybean Board (USB), National Corn Growers Association (NCGA), Western Growers Association (WGA), International Fresh Produce Association (IFPA), university and USDA ag extension services, independent crop consultants, cooperatives, and state/regional farm bureaus is key to promoting wider-spread adoption.

5 Having a Playbook

The final difference is that testing and bringing a pharma product to market usually follows a well-known playbook. Hit finding, optimization, ADMET tox, pre-clinical tissue & mouse, larger mammal, Phase I, Phase II, Phase III, and Phase IV trials are understood in pharma product development. Government websites even summarize this [2]. The power of these studies to indicate the stage of product development is acknowledged by investors and entrepreneurs alike. AgTech has lacked a similar common understanding of the stages of its testing and development pipeline. This is due in part to R&D operations and strategy being considered a competitive advantage in bringing products to market, with asymmetric information leading to company valuation differences, as well as the ever-evolving AgTech regulatory landscape.

Even with an established Playbook, pharma product development faces large upfront costs with ranges between $500m and $2B for developing a single drug [3] and significant risk with only about 12% of drugs moving from starting clinical phase I trials all the way to market [3].

AgTech product discovery and development is similar. While the regulatory path and the types of testing are different, there is nonetheless also a long, expensive, and high-risk path to market. Our goal is to demystify that path. Like all playbooks, the expectation is that this document will change and adapt over time as it is adapted to provide guidance towards having evermore successful product pipelines. This document is not meant to be prescriptive, but rather to provide a framework and shared nomenclature around the stages of development for key product classes in agriculture.

The Playbook aims to bring transparency and a shared nomenclature of R&D in agriculture to aid entrepreneur and investor alike in bringing new innovations to the AgTech sector.

The Six ‘R’ Eras of AgTech

New Chapter

Author:
Scott Porter

Special Thanks:
Adam Bergman

Agriculture has always been foundational to human civilization. It provides the food, energy, and raw materials that sustain population and enable economic development. Beyond simply feeding the world's 8.2 billion people, agriculture underpins the global economy (representing ~4%, or ~$4 trillion, of global GDP[2]), creates jobs (on-farm agriculture employs ~28%, or ~1 billion, of the world's working population[3]), and drives global trade (representing ~8%, or ~$1.7 trillion, of global trade[4]). As global demand for food continues to rise alongside population growth and shifting consumer preferences, the importance of agriculture continues to increase.

Yet the farmer is facing unprecedented challenges – water scarcity, labor shortages, climate and environmental risk, disease resistances, soil degradation, regulatory burden, and economic uncertainty, to name a few. However, like in many sectors of the economy, AgTech innovations can overcome existing challenges and drive future progress.

This backdrop combined with the opportunity provided by growing commodity prices after 2020 attracted capital markets to the AgTech sector, but in 2024, AgTech saw a decline of more than 50% in fundraising activity from its peak in 2021[5] and this trend has continued into 2026 To understand this pullback, and the path ahead, we must first examine how we arrived here.

It's worth noting that AgTech can mean many things – for the purpose of this capital markets overview, it's best to bifurcate the market into two buckets: the regulated market (requiring government agency approval – the scope of this Playbook) and the unregulated market (e.g., automation, robotics and digital products).

How We Got Here – The First Four 'R' Eras

The Recondite Era (2005–2013)

Esoteric, obscure, and overlooked. AgTech in this period was little understood or appreciated. Startups were few, funding was sparse, and dedicated AgTech funds and Corporate Venture Capital (CVC) arms like Syngenta Group Ventures existed but were rare with the 'Big 6' ag firms focused on developing plant biotechnology solutions. Generalist VC interest was minimal, leaving most companies to bootstrap or operate with modest resources. Yet many of the boldest AgTech innovators quietly took root in this era, building with less capital, lower expectations, and more time.

The Rational Exuberance Era (2014–2019)

The ~$1B sale of the Climate Corporation to Monsanto in late 2013 signaled the arrival of AgTech, ushering in the sector's first unicorn and a flood of capital. An increase in the number of dedicated AgTech venture firms quickly followed – S2G Ventures, Lewis & Clark Ventures, Pontifax AgTech (now Aliment Capital), Anterra Capital, AgFunder, and iSelect all launched between 2013 and 2014. Soon after came a wave of CVCs: Leaps by Bayer and Yamaha Motor Ventures (2015), ADM Ventures (2016), Bunge Ventures and Cavallo Ventures (2017), and Kubota Innovation Fund (2019), to name a few. This surge of awareness and capital sparked a boom in entrepreneurship: the era averaged 1263 new AgTech company formations per year, versus just 271 annually during the Recondite Era[6].

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[2] Source: World Bank Group: https://data.worldbank.org/indicator/NV.AGR.TOTL.ZS

[3] Source: International Labour Organization: https://www.ilo.org/topics-and-sectors/industries-and-sectors/agriculture-plantations-other-rural-sectors?utm_source=chatgpt.com

[4] Source: https://www.wto.org/english/tratop_e/agric_e/ag_imp_exp_charts_e.htm

[5] Source: PitchBook, Cascadia Capital data & analysis

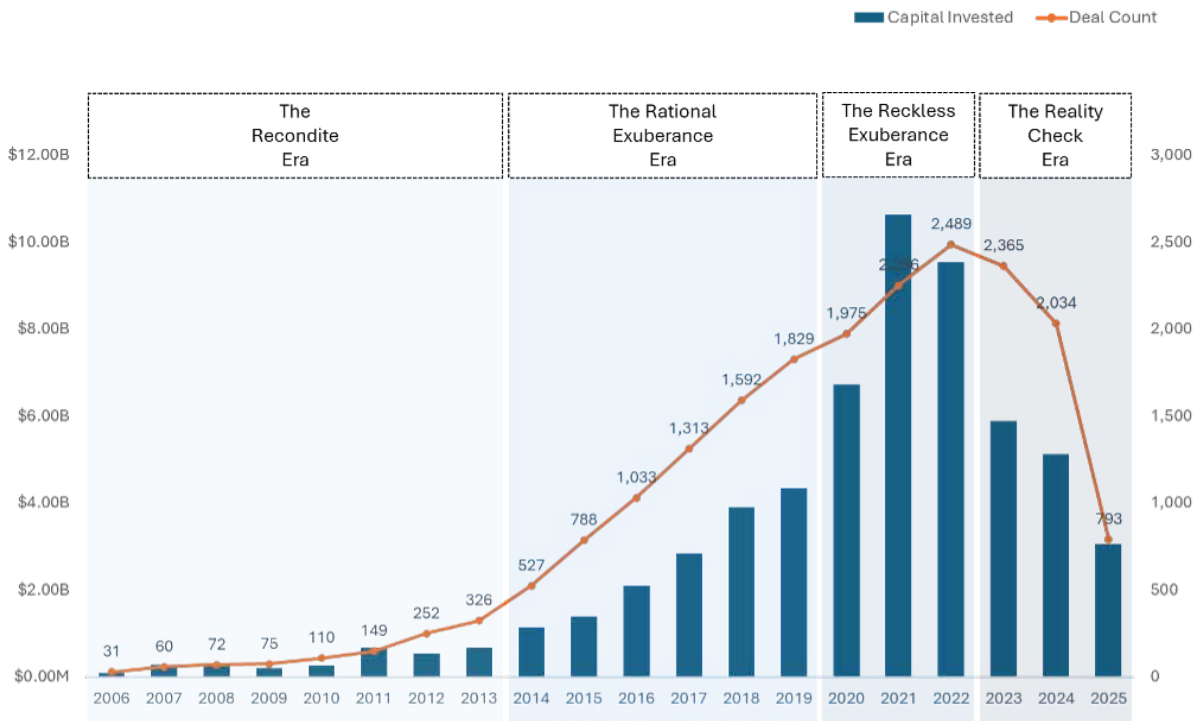
The Reckless Exuberance Era (2020–2022)

Low interest rates, abundant capital, growing sustainability and impact focus, and peak AgTech optimism fueled soaring valuations and oversized rounds. Generalist funds who previously shied away from AgTech, piled in – Andreessen Horowitz, Google Ventures, SoftBank, General Atlantic, and TPG among others. Entrepreneurs embraced a "grow at all costs" playbook and were rewarded with rising valuations. Median post-money valuations more than doubled from the Rational Exuberance Era, while annual capital invested nearly quintupled[6]. AgTech did see a number IPOs or special purpose acquisition companies (SPACs) occur this period, though valuations rarely help up in the public market.

The Reality Check Era (2023–2024)

Rising interest rates, commodity volatility, supply-chain shocks, and extreme weather forced a pivot toward demonstrable ROI. Most telling, the Playbook's realities – multi-year field trials, regulatory hurdles, and costly scale-up – proved immutable despite advances in computational discovery. The beginning of valuation resets through down rounds and restructurings, as companies missed milestones, liquidity tightened, and bridge rounds and extensions became the norm. The true length and complexity of the AgTech sales cycle came into sharp focus.

AgTech Investments & Deal Count[7]



[6] Source: PitchBook, Cascadia Capital data & analysis

[7] Source: Pitchbook. Note: Represents global transactions. 2025 is year-to-date.

Two ‘R’ Eras to Define the Future of AgTech

The Reckoning Era (Current)

Bridge rounds and extensions have run their course. Companies must now prove themselves in the capital markets, creating a “winners and losers” market. Winners will adapt to current market conditions and secure growth capital, whereas others will face down rounds, restructurings, or bankruptcy. Although capital remains available for milestone-ready players (e.g., Carbon Robotics, Pairwise, Ever.ag), this period will be defined by mixed signals: more invested capital and exits, but also a continuing painful shake-out.

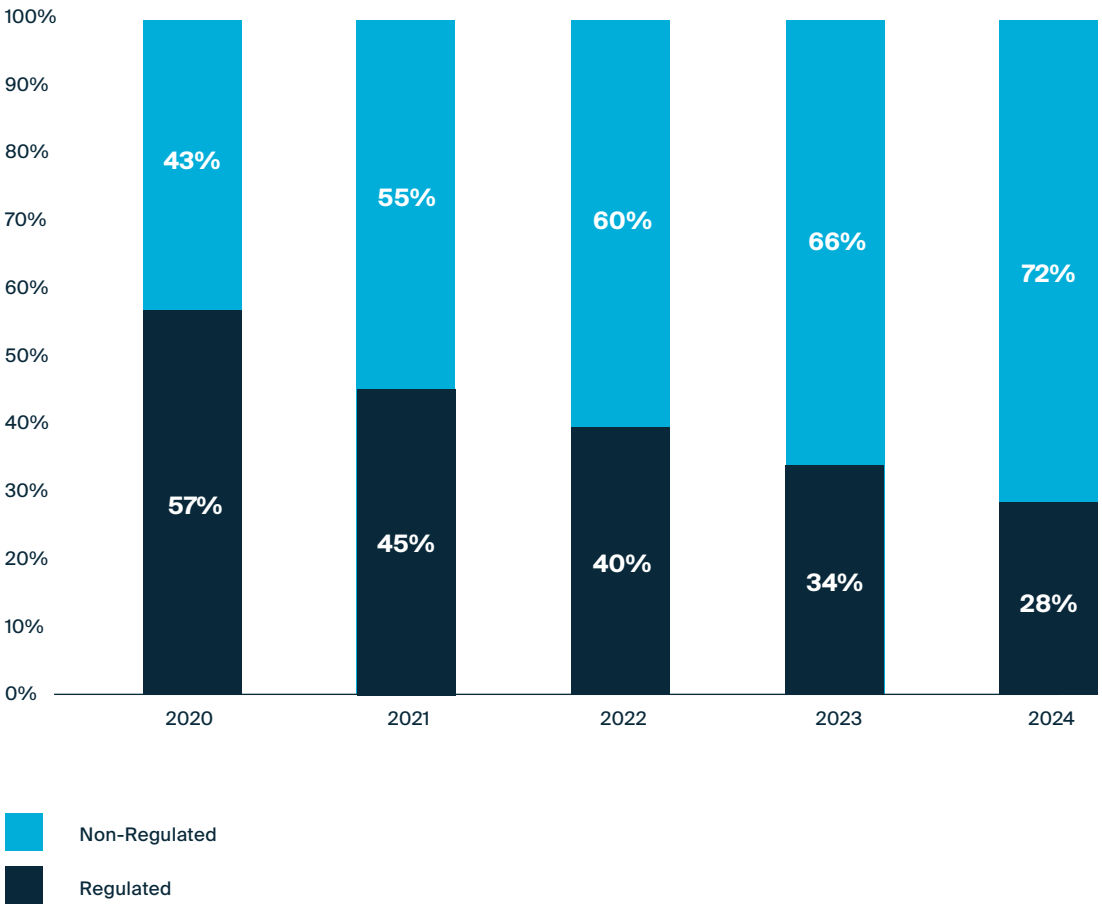
The Renaissance Era (Future)

Capital volumes experienced in the Reckless Exuberance era may not return for a long time; however, AgTech returns to rational exuberance grounded in a clearer understanding of the AgTech timeline and the true cost required to bring innovation to market. This period will be marked by fewer deals and lower valuations, but higher quality and more market wins. More examples of commercial traction from earlier-generation startups will exist in the market as models for Renaissance founders to follow. Entrepreneurs and investors embrace longer timelines and balanced expectations, paving the way for game-changing technologies and durable exits.

Fundraising and the “Valley of Death”

‘Where have all the investors gone?’ has become the sector’s most common refrain. Consistent with what has happened in other sectors (solar, electric vehicles, etc.), as battle scars mounted, many generalist investors exited AgTech, while specialist funds shifted either earlier (seed) or later (buyout/private equity). These funds can struggle to raise additional capital to deploy until they can show success and return of capital from earlier investments. Investor caution has sharpened around regulated AgTech, where long timelines and unproven milestones remain barriers for new capital. For the first time, Precision Ag is outpacing Ag Biotech in funding, a reflection of investor preference for faster commercialization in unregulated markets. The result is a widening Series B–C funding gap, particularly for regulated companies: AgTech’s “Valley of Death.”

Regulated vs. Non-Regulated AgTech Funding as a % of Total Investment[8]



In this winners and losers environment, many companies will not survive. Yet the coming Renaissance Era is likely to bring a new class of long-term capital that will drive this next era's success – family offices, CVCs, pension funds, sovereign wealth funds, and long-term purpose-built vehicles focused on scaling best-in-class regulated technologies for regulated markets.

[8] Source: PitchBook, Cascadia Capital data & analysis

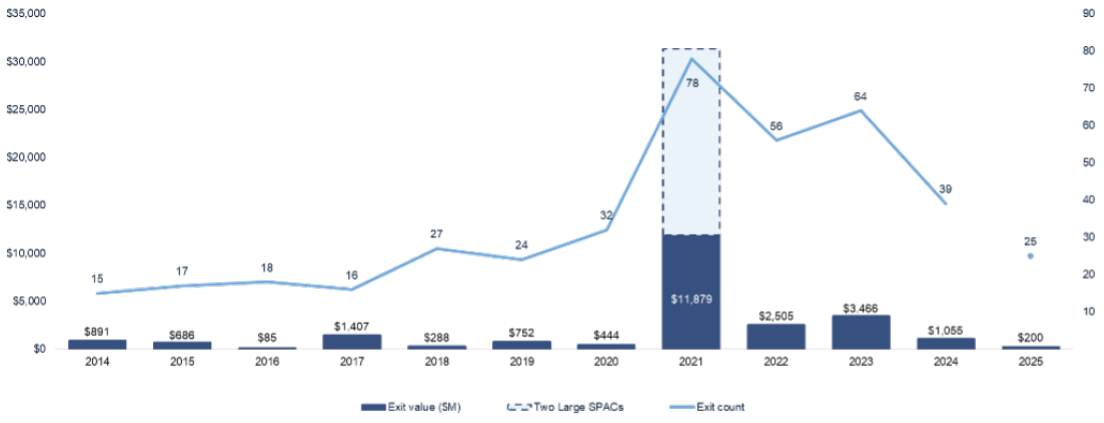
M&A and the Exit Conundrum

At the heart of AgTech's financing challenges is the lack of exits. The sector has returned little capital to date and even 2021's apparent "peak" was misleading, with Ginkgo Bioworks and Benson Hill going public via SPACs but returning little true capital. Strip those out and exit activity has consistently fallen short of the hype. Strategics remain interested, but unwilling to pay 2021 level valuations, while most AgTech fundamentals are not yet ready to attract public market investors or private equity buyers.

Yet, even though exit volume is tracking higher in 2025, exit value is down considerably, which can be divided into four categories:

1	**Distressed M&A**	IP or asset acquisitions at floor prices.
2	**Early M&A**	Companies that have yet to, or will not over-raise and can deliver acceptable returns at valuations strategics will pay.
3	**System Add-On M&A**	Subscale companies that integrate seamlessly into incumbent portfolios.
4	**Old World "Tech-Enabled" M&A**	Cash flowing, family-owned or self-funded businesses leveraging technology outside of the VC markets.

AgTech VC Exit Activity[9]



Ultimately, it is still too early to judge. With time-to-market measured in 4–13 years, many regulated AgTech companies remain in their infancy. But one thing is certain: the next 5–7 years will be decisive in proving whether AgTech can consistently deliver real exit opportunities.

[9] Source: PitchBook and Cascadia Capital analysis

Recommendations for Navigating the Capital Markets for Investors, Entrepreneurs, and Farmers

AgTech remains critical to humanity, but success in the Renaissance Era requires learning from past capital markets missteps. Three imperatives stand out:

Investors	*'That's great, but I want you to tell me how you're going to put a cow on the moon'* suggested one investor. Venture capital has thrived on moonshots, but AgTech demands patience. In the rush for growth, investors pushed entrepreneurs to over-promise relative to the long timelines and heavy costs of regulated innovation. Going forward, disciplined underwriting and a return to rational exuberance are essential. Valuation discipline will enable better returns of investment and success for all parties.
Entrepreneurs	As one seasoned investor put it, *'Dilution never killed anyone, but too much capital has.'* Over-raising has doomed promising companies with bloated cap tables, unsustainable burn, and exit hurdles too high to clear. The path ahead requires long-term discipline: take only the capital you need, and above all, listen to the farmer, not just the investor. Partnering with the ecosystem unlocks long term growth and can put less of a capital burden on companies.
Farmers	True success requires skin in the game. Like all consumers, farmers cannot always predict what technology/solutions they will want in the future. However, farmers must be willing to invest time, effort, and resources into testing and adoption. Without their engagement, no technology – no matter how well funded – can scale.
Limited Partners	Know what industry you are getting into. The return profile of funds and companies in AgTech can extend beyond the standard return cycle expectations of a venture fund. The track-record of the funds and the experience of the General Partners in AgTech should be critical considerations.

The Critical Role of Regulatory Strategy

New Chapter

Author:
Jerry Hjelle

Special Thanks:
Eric Park

Ag Playbook 2.0
Thinking Regulatory Strategy

Disclaimer: Regulatory strategy and the laws and entities surrounding it, constitute a constantly evolving landscape. This section focuses on a range of crop-protection products mentioned elsewhere in the Playbook and is not 'all inclusive'. While we hope you find this section informative, it is advisable to seek the help of experts in the field to best plan your path of product development and approval.

Obtaining regulatory freedom to operate is an important milestone in AgTech product development and commercial success. Whether the project involves a new small molecule, biomolecule, or a biotechnology plant, the associated testing, approval, and stewardship requirements are often complex, expensive, time-consuming, and highly variable due to external governmental delays. In most complex cases of biotechnology commodity crops, current rules regarding adventitious presence of recombinant DNA (rDNA) create the expectation for multiple, coordinated data packages, dossiers, and approvals in key export markets. By contrast, if the product is a biomolecule pesticidal protein or double-stranded RNA (dsRNA) with low probability of residues in the finished food or feed, when used locally under conditions of intended use, then markets can be prioritized and sequenced without the need for approvals in global export markets. This section will focus on engaging government bodies and not specifically on certifying organizations e.g., non-genetically modified organism [non-GMO] label) that may require separate data to apply the associated labels.

The AgTech Playbook 2.0 concept is critical because decentralized *technology innovation* is increasingly separated from *product development*, *regulatory,* and *market-launch* expertise. Regulatory strategy often plays the advising role of guiding technology innovation companies to define the product, its intended uses, and target markets. An integrated regulatory strategy creates competitive advantage as it enables smooth transitions across product development phases and full lifecycle. As shown in the **Six 'R' Eras of AgTech** section, investors are now shying away from projects considered to be 'regulated' AgTech. This generalized aversion is an opportunity for startups and investors who are knowledgeable regarding regulatory pathways and de-risking strategies.

The AgTech Playbook 2.0 stage-gate structure is intended to both *drive organizational alignment and enable disciplined portfolio risk/resource management decisions*. For regulatory, Phase 1 should consider the "theory of approvability" of likely products. This includes thinking through the uses, processes, and end markets to identify the "who and how" of needed approvals while considering opportunities and obstacles. Phase 2 involves the development of a creative regulatory strategy and supporting studies, as product type and process will influence agency classification, data requirements, and review timelines. Although specific guidance often exists, consultation with regulatory agencies can clarify data requirements, especially for novel products that do not align neatly with existing regulatory pathways. Experienced regulatory experts recognize that each new product, intended use, and target market represents a unique case, and that a strong understanding of risk-assessment principles (exposure, hazard assessment, history of safe use, bioinformatics) supports productive discussions with regulators regarding data requirements and potential waivers. **Proactive regulatory action during Phase 2 can reduce expensive later-stage failures by conducting de-risking safety and environmental screening studies.** Startups seeking to license their technology to or be acquired by an incumbent prior to Phase 3 may face unexpectedly rigorous regulatory due diligence and valuation discounts if their regulatory plan and risk mitigation strategies are inadequate.

Entering full-scale Phase 3 regulatory testing represents a significant resource commitment. Testing can include process definition, five-batch production, analytical methods, specification

setting, formulation development, toxicology, ecotoxicology, and environmental fate studies. **At this stage, changes in product specifications, process, use, or formulation, may force new studies and create regulatory delays—the so-called regulatory lock-in.** Lock-in refers to all ingredients, including "inert" ingredients that can similarly impact the regulatory review process. Testing protocols should consider key market requirements, even though global dossier submissions may be staggered. Regulatory consulting groups can help manage agency pre-consultation meetings, testing, contract research organizations, dossier production, and approvals.

Cost and Timing in Global Regulatory

Regulatory costs and timing can vary significantly depending on the type of AgTech product. Generally, biomolecules that resemble naturally identical structures have an advantage compared with new small-molecule chemical and biotechnology crops traded globally.

For example, regulatory costs for large companies (CropLife, 2024) ranged from US$42–52 million for small-molecule active ingredients (AIs) and large row-crop biotechnology traits with approval timeframes often spanning 5–7 years, driven largely by the lengthy review process and highly prescribed studies in the EU and China. By contrast, the costs and timelines for approval of purified insecticidal biomolecules, often produced by fermentation of genetically modified (GM) microbes, are US$1–7 million in the US and Brazil across 18 months to 5 years. Unlike biotechnology traits crops such as plant incorporated pesticides (PIPs), approval of biomolecule biopesticides in specific markets can enable market entry one at a time if residue levels in exported commodities sprayed with the biopesticide under conditions of intended use are not detectable, or a d*e minimis risk*.

Generally, the safety requirements for biomolecules (e.g., insecticides, fungicides) must result in the same US Environmental Protection Agency (EPA) conclusion as with small molecules and PIPs. The applicant must show that the product will not generally 'cause unreasonable adverse effects…' to human health and the environment under conditions of intended use. For small molecules, with no previous precedence of testing or approvals, a broad array of expensive and time-consuming studies is required to identify potential health and environmental hazards, assess environmental fate and exposures, and characterize risk. By contrast, sprayed purified biopesticide classes (e.g., proteins, dsRNA) with a history of safe use/exposure or low exposures and highly selective toxicity may qualify for significant data waivers. The good news for developers is that several regulatory agencies, for example in the US and Brazil, are open to, and often welcome, pre-consultation meetings where individual product information can be discussed. These regulatory agencies are also open to case-by-case arguments supported by the appropriate data. Agencies, for their part, must examine ways to streamline guideline-required studies, especially for products with very low probability of harm products (common biomolecule classes) and low/no food or feed exposure situations.

Entrepreneurs and research & development (R&D) leaders often consider regulatory activities as a glorified box-checking exercise. In reality, it is the process of establishing that your product is safe and efficacious under the conditions of intended use. Due to the importance of conducting and reporting health and environmental safety studies and legal disclosure requirements of regulatory authorities (e.g., US EPA, US FDA [Food and Drug Administration], IBAMA [Brazilian Institute of Environment]) it is also important to understand the significant levels of documentation needed for these regulatory activities. Specifically, key studies are required to have defined protocols and documentation that reflects any deviations and amendments. Often, quality assurance professionals

are needed to assist startups in recording production lot standard operating procedures and sample chain of custody, which are two cornerstones of regulatory science. Stated differently, the research mindset must incorporate a regulatory data production focus, with the regulator's perspective: *if you didn't document it fully, then it didn't happen*. Typical R&D studies do not meet the authority-recognized study design and methods, thus involving regulatory experts could lead to time and cost savings in bringing a product to market.

The complexity of integrating regulatory considerations into product development can seem daunting. However, it is possible to build a regulatory scorecard for a product/class that tracks along with the overall development plan, facilitating communication and action for R&D, management, investors, and stakeholders. There are key milestones along the path of regulatory product testing, submission and approvals; regulatory plans and outcomes should be managed like any other highly important critical path program.

Example Case for Biomolecule Biopesticide Regulatory Process in the USA and Brazil

Let's examine how a specific biopesticide would be regulated in two key potential markets: the USA and Brazil. These markets were chosen based on their size, general clarity on the guidance, and openness to both pre-submission consultations and data waivers considering a case-by-case approach, depending on the intended use of the product.

Example Product Specs

Product type	Biomolecule-based bioinsecticide for in-season use in major row crops (e.g., corn, soy)
Production method	Produced through fermentation and downstream processing using a GM microorganism recognized as safe, with representative examples in the USA and Brazil.
Detection and residue profile	Neither the AI nor formulated product contain detectable living modified organisms, but the rDNA and newly expressed protein or dsRNA are detectable using standard, accepted methods used by the industry and regulators. Use of the formulated product, under conditions of intended use (spray rate, harvest interval) does not produce detectable residues of the biopesticide on either the finished food or feed commodity.

United States

GM microbial–derived insecticidal proteins and dsRNA products that are nonliving and intended to control pests are regulated as **pesticides**. As such, they are regulated by the US EPA under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) and the Federal Food, Drug, and Cosmetic Act (FFDCA). FIFRA grants EPA the authority to allow sale and distribution and FFDCA grants EPA the authority to establish legal limits for residues in food or feed, or to determine that a limit is not needed (exemption from the requirement of a tolerance). Because the technical grade active ingredient (TGAI) is a semi-purified product and does not contain viable GM microorganisms, the United States Department of Agriculture's Animal and Plant Health Inspection Service (USDA's APHIS) typically has no regulatory role unless a living genetically engineered microbe or plant is being released into the environment. The FDA is only involved to enforce pesticide residue tolerances that the EPA sets for products used on food crops for animal or human consumption. For dsRNA, the EPA also evaluates sequence specificity, off-target potential, and environmental stability, as outlined in its RNAi-focused guidance. Because the TGAI and formulated products are classified as biopesticides, the Biopesticide and Pollution Prevention Division (BPPD) within the EPA will take the lead in reviewing and approving the product. This includes the Section III approval of the use of the biopesticide and a decision on the exemption or definition of tolerance level on residues in food or feed. The BPPD has defined specific categories of products, including microbial and biochemical biopesticides. These include guidelines for specific information to ultimately approve the product for intended use with accelerated timelines and a reduced fee structure (compared to small-molecule pathways).

The external regulatory process begins with a pre-submission consultation with EPA-BPPD. During this, the specifics of the TGAI and formulated product, including intended uses, are presented for the case under review at the end of Phase 2 or beginning of Phase 3. Prior to this meeting, a clear presentation and strategy should have been developed, supported by data and/or literature, regarding the safety of the TGAIs, the pattern of use and exposures, presence on commodities, etc. The goal is to make the case for the classification of the product, in this instance, as a biochemical biopesticide. By knowing the information and data needs for this category, arguments are made for waiving specific biochemical biopesticide data requirements by using published or generally known information. This meeting will clarify possible data waivers and identify any new data needs.

This is followed by submission of a FIFRA registration package, which includes a combination of literature, precedence or new information on product chemistry, toxicology, ecological effects, environmental fate, and exposure data. Although a full registration is still required, the data package is more streamlined than for living microbial pesticides because nonliving materials cannot replicate or cause infectivity; thus, the EPA does not require microbial identity, viability, pathogenicity, or colonization studies. Instead, the risk assessment focuses on product chemistry, acute toxicity, nontarget organism effects and environmental degradation, with additional consideration of sequence specificity and off-target potential for dsRNA. The EPA has a separate data requirement for biochemical (biomolecule) products that allows for customization of studies needed for registration.[10] The EPA then conducts a comprehensive human health and ecological risk assessment, and if the product is used on food crops, it also establishes a tolerance or exemption under FFDCA. Once the review is complete, the EPA issues a registration decision with approved uses and labeling, serving as the central authority ensuring safety and efficacy before the product enters the US market. Under the applicable EPA Pesticide Registration Improvement Act review categories for this case (B590), the typical review timeline is approximately 20 months. However, the end-to-end process may reasonably extend to 20–30 months.

The above regulatory activity is at the federal level. Please note that state level registration is also needed. Approved inert ingredients will not generally trigger regulatory issues.

[10] https://www.epa.gov/test-guidelines-pesticides-and-toxic-substances/series-880-biochemicals-test-guidelines

Brazil

The regulatory pathway for a GM microbial–derived insecticidal protein and dsRNA product is more complicated in Brazil. Brazil's regulatory framework for GM microorganisms involves multiple agencies working in coordination. For GMOs specifically, the National Technical Biosafety Commission (CTNBio) handles biosafety approval and the classification of the product. The Ministry of Agriculture, Livestock and Supply (MAPA) issues the final registration certificate. The National Health Surveillance Agency (ANVISA) under the Ministry of Health reviews health and toxicology aspects, while the Brazilian Institute of Environment (IBAMA) conducts environmental review.

The process for GM derivative products begins with the classification of the product as a GM derivative or non- GM, and in some cases, CTNBio biosafety approval of the microorganisms.

Highly purified biopesticides can be exempted from CTNBio review under specific circumstances. Products obtained by gene-edited organisms can also be classified as non-GM in a case-by-case analysis by CTNBio. To understand these circumstances, it is important to understand that the Brazilian Biosafety law (11.105) classifies products as:

GMO	An organism with genetic material (DNA/RNA) that has been altered using any genetic engineering technique.
GMO derivatives	A product derived from a GMO that does not have the ability to replicate independently or does not contain a viable form of GMO.
Exclusion from GMO/derivative category	A pure and chemically defined substance obtained through biological processes is not classified as a GMO derivative if it does not contain a GMO, heterologous protein, or recombinant DNA. Purified GM protein is also classified as non-GM if it is highly purified.

In addition, CTNBio created Normative 16, which ensures proper regulatory classification of emerging gene-edited products and included RNAi in the scope. It is a case-by-case consultation system, designed to assess whether a product should be considered a GMO. In this case, CTNBio will evaluate mainly how the product was obtained and its application to define the classification. When a product is classified as a GMO, it must undergo CTNBio's comprehensive biosafety risk assessment prior to approval. If the product fits into one or more of the Normative 16 criteria, products can be determined as non-GMO, and may be registered through existing regulatory procedures determined by MAPA, ANVISA and IBAMA, with no need for additional biosafety evaluation.

Following a strict regulatory pathway, GMOs in Brazil must undergo biosafety approval by CTNBio prior to other specific registrations and commercialization. This includes authorization for field trials with regulated GMOs, which are categorized as "Planned Release into the Environment" and are conducted under controlled conditions. CTNBio's risk assessment focuses on biosafety issues such as potential effects on target and non-target organisms, soil microorganisms, and other features of the organism's biology. If the product is classified as non-GM, it will go directly to the registration process with no need for a biosafety evaluation. In this case, the authorization for field trials by CTNBio is exempted.

Following CTNBio approval, the product must go through the pesticide registration process, which involves parallel review by three agencies. Applications must be submitted simultaneously to MAPA, ANVISA, and IBAMA. The data package must include information on physico-chemical or

biological properties of the microorganism, toxicological studies, and ecotoxicological studies. Notably, toxicological studies can be conducted in other countries and are accepted for registration purposes as long as they are carried out under good laboratory practices. However, efficacy trials must be conducted in Brazil—applicants must present three field trials per target and/or crop in three different regions, though only one season is required.

The question of whether a technical product registration is required for microbiological products is examined on a case-by-case basis. For many biopesticides, TGAI registration is not required, though certain information on the AI must still be provided.

During the review phase, MAPA evaluates efficacy, ANVISA reviews toxicology and health aspects, and IBAMA assesses environmental impacts. Upon completion of all three reviews, MAPA issues the registration certificate as the lead regulatory authority.

The timeline for approval in Brazil varies dramatically depending on product type. The average time for approval of a generic formulated chemical product is 5–6 years, while in the case of low-risk products (which includes biological and microbiological products), this period is only 14 months. However, for new GM microorganisms, the total timeline including CTNBio approval may extend beyond four to five years. The exception is non-GM classification through Normative 16, which can decrease CTNBio's timeline to 2 to 6 months.

Brazil has made significant strides in recent years to encourage biopesticide development. The 2020 Decree 10.375 simplified the registration process for biopesticides and biofertilizers. More recently, the 2024 Bioinputs Law (No. 15070/2024) provides a comprehensive new framework for biological products, though further regulation is still required. Brazilian legislation has been taking large steps in the regulation of biopesticides to specify requirements and reduce the period for approval of registration.

Thinking about the production method, in Brazil, multi-functional microorganisms that possess both pest control and growth promotion properties require multiple separate registrations for different uses. For example, a *Bacillus* strain might need to be registered separately as a growth regulator, a biopesticide, and a soil conditioner, leading to redundant data submissions. However, Brazilian authorities are working to address this, for example by constructing an interdepartmental database and encouraging compound product development.

Proactive Advocacy for Improved Global Regulatory Pathways for Biomolecules for AgTech

The US EPA-BPPD and Brazilian regulatory authorities are currently looking for and implementing ways to streamline regulatory processes for the approval of biopesticides, using principles of history of safe use and familiarity/similarity to established safe proteins and RNAs. In the vast majority of situations and uses, these biomolecules have a low probability of harm to health and the environment with a low likelihood of persistence in the environment and low/no human exposures. They understand the case-by-case uniqueness for regulatory problem formulation and support the use of pre-consultation meetings to improve communication, alignment and resource efficiency. Leaders should advocate to improve the approval processes and transparency in target markets to unlock the use of AgTech biomolecules in numerous other markets that have yet to streamline and revise their guidance and regulations.

What's in the Playbook and What's not?

The Playbook is designed to help founders and investors better understand product development. The first two chapters focus on novel AgTech product development and illustrate the types of experiments, trials, and results that investors and regulators may expect from entrepreneurs and companies.

These guidelines are not hard-and-fast rules and do not necessarily correspond to a startup's funding stage (e.g., Series A, Series B), since a company may have multiple products at different stages of development. A rough estimate of the costs associated with conducting these experiments is provided to support planning and fundraising strategies. This Playbook is not all-inclusive, and regulatory requirements are likely to change over time and vary by country and state. Startups should consult the appropriate regulatory agencies and experts early in the product development process to ensure they are planning appropriately and are compliant with applicable regulations and laws.

The Playbook does not address go-to-market strategies or novel innovation systems for conducting research and development (e.g., shared infrastructure for field testing); rather, it focuses on the specific types of work that need to be accomplished to develop next-generation products for farmers.

The Playbook
Executive Overview

Developing AgTech products is a long and challenging process, requiring the intersection of multiple scientific disciplines.

The first two chapters of this Playbook will look specifically at what it takes to research and develop **crop protection products**. Future chapters may focus on other forms of crop improvement technologies, biological products, digital ag products, and animal ag products.

Each type of product comes with its own challenges in the discovery phase, but all products need to go through similar development **Scale-up** and **Field & Regulatory Trial** stages that are ultimately subject to seasonal dependence.

Across product categories, meeting regulatory requirements and product concept standards while proving efficacy and safety in broad acre field trials can be challenging. Conducting field trials are often the costliest and longest part of bringing products to market. Generally, there is only one spring planting and fall harvest season for each Northern and Southern hemisphere growing region, and time-to-market will depend on successfully executing the required field trials. This is excluding the multi-season growing regions found between the latitudes 10 degrees north or south of the equator that can be used to accelerate timelines as they can have more than one growing season in a calendar year. In agriculture, delays and setbacks are not measured in weeks or months but years. Ultimately weather and environmental conditions can never be completely anticipated or controlled. This challenge further emphasizes the importance of ensuring proper protocol design to power analysis at the end of any season.

Due to the diversity of AgTech product types and how they are regulated, it is difficult to generalize a single timeline or path to market. Some **biofertility** products may have a seemingly quick route to market, taking as little as 5 years, but it can be difficult to discern actual product impact distinct from other environmental impacts. **Genome editing** has the potential to provide a more rapid route to market than genetic modification (GMO) technologies, but crop plants with the genome edits still need to be field-tested and introgressed into commercially relevant germplasm to demonstrate the added value. Novel uses of artificial intelligence can decrease the duration of hit finding and optimization steps of **small molecule or biomolecule discovery**, but formulation work and the volume and scale of required field trials expands the later stage costs and extends the time of development.

AgTech products strive to show consistent performance in an ever-changing world and the hope is this that Playbook serves an aid in the development of those products that improve farmers and consumers lives.

Each product pipeline faces its own challenges but there are a few consistent truths here to emphasize:

1

It takes time (3-13 years) to bring any of these products to market.

While more money can be raised, more time cannot be created. Having rigorous planning around the seasonality of agriculture, including field and registration trials/studies, is critical to success.

2

It takes significant capital to develop AgTech products.[11]

Testing in plant model species can begin rapidly for most product types. However, the capital cost to field test in crop plants at the diversity of locations needed to prove efficacy and crop safety continues to rise. And this is only about the path to develop a product; more costs will come to maintain and grow a product in the commercial market.

3

There is a high burden of testing to prove these products will work well within the integrated system of existing agricultural technologies and practices.

Claims of increasing yield using genome editing technology will need to be validated in commercially leading germplasm in large scale, multi-location field trials. A new crop protection small molecule product will need to fit into a system and avoid antagonistic effects with other commercially applied chemistries to be widely adopted. A biological that has a 65% win-rate is only slightly above random chance and therefore not likely to bring the value necessary to move the skeptical consumer.

4

Working with partners and farmers is key to building *belief* and establishing consumer trust in the market.

As mentioned in the earlier problem section, often in agriculture we see the Reverse Field of Dreams: Just because you build and bring a product to market, does not mean the customers will necessarily come. Conducting field trials with trusted third-party partners and farmers and then publishing these results will support claims of product efficacy. By partnering to conduct trials with farmer engagement groups, test farms, universities, retailers, and long-standing companies with strong product histories, innovators can build a stronger base of trust in the results they are reporting.

[11] Software products may be developed quickly and at lower cost, however, will still be subject to testing, adoption, and integration with existing systems.

Phases of Ag Products
Executive Overview

Phase 0
Product Concept

Phase 0 is the starting point where you will define the problem to be solved, the size of the opportunity, and current solutions to the problem (later this will be your positive control group!). Taking the adage to heart: "If I had an hour to solve a problem, I'd spend 55 minutes thinking about the problem and 5 minutes thinking about solutions." The same mentality should be applied to this phase as it can save time and money down the line. This phase should be heavy in interviews of farmers/customers of a future product.

Examples:

In the case of crop protection small molecule products, this phase includes defining the intended crop, pest/pathogen, and molecular target of interest and understanding the current product offerings and unmet need in the market.

In the case of digital ag hardware devices, this phase includes establishing the baseline for current measurement, labor, or operational practices.

Phase I
Pre-Field Discovery

Phase 1 or the "Pre-Field" phase of product development focuses on demonstrating the efficacy of a novel lab or technology discovery. This often includes screening candidates and advancing leads through rounds of iterative trials, redesign, and testing. This is the proof-of-concept phase where the focus is on identifying the lead candidate or product design and testing it against the product concept. Lead candidates should undergo an initial assessment of any prohibitive safety concerns and researcher should prepare a regulatory engagement and approval strategy. Exiting Phase 1, a product should clearly demonstrate high efficacy under controlled conditions and clear differentiation from existing solutions on the market.

Examples:

In the case of crop protection small molecule products, this would include molecular and pathogen target refinement, candidate screening, hit discovery, structure database and patent search review, and lead optimization for new active ingredients.

In the case of a digital ag hardware device, it would mean prototype development and testing.

Phase II
Early Product Development

This phase focuses on three elements: 1) conduct further assessment of any safety concerns and execute the initial regulatory engagement and approval strategy, 2) review leads for novelty and freedom to operate and implement an intellectual property (IP) protection strategy, and 3) demonstrate consistency in performance in a broader diversity of conditions and locations.

Examples:

In the case of crop protection small molecule products, this would involve completion of ecology and toxicity database screens, formulation development, initial cost of goods assessment, and efficacy proof in growth chambers, green-houses, and small plot field trials.

In the case of a digital ag hardware device, it would mean patent review, and movement from prototype to initial product testing.

Phase III Advanced Product Development

Phase III focuses on refining and executing the regulatory approval strategy, generating the required data for regulatory dossiers, and refining the product concept. Many studies at this phase will need to be executed at certified facilities that comply with testing standards recognized by regulatory authorities such ISO standards, GLP[12], and GMP[13]. Phase III includes improving the cost of production for scale-up, achieving key product safety milestones, and demonstrating widespread efficacy. It also generally includes an increase in the reps and scale of field trials at a wider diversity of locations to confirm product efficacy data generated in earlier phases. Engaging legal and regulatory experts early and often is critical during this phase to ensure freedom to operate and value capture for the product upon launch. In addition, early engagement with farmers/customers and affiliate organizations can lead to much easier Phase IV and Phase V experiences.

Examples:

- In the case of crop protection small molecule products, this includes scale-up production of active ingredients and inerts, wider testing in small and large acre field trials, regulatory trials and testing, and advanced formulation evaluation, including packaging and related long-term product stability.
- In the case of a digital ag hardware device, it would mean taking the product out to the field for side-by-side testing against the best-in-class standard.

Phase IV Pre-Launch Preparation

This phase will focus on supporting the data packages and dossiers being evaluated by regulatory authorities as well as preparation for the upcoming product launch. Simultaneously, a broad data set should be generated to support future marketing and sales efforts. The focus should be not only on building up the data set but also on implementing a literature publication strategy, creating a product stewardship strategy, and developing an effective go-to-market plan.

Examples:

- In the case of crop protection small molecule products, this requires a technical profile for the new active ingredient to be fully defined, that a core formulation concept is established and is free for registration, and that the global dossier to regulatory authorities for the initial key market(s) are complete.
- In the case of a digital ag hardware device, it implies widespread testing of the hardware in various regional, soil, weather, and time conditions to ensure its broad use. This is also where workflow analysis can best be tested.

Phase V Launch and Market Expansion

This launch phase focuses on continual testing to aid the business development and expansion into new markets as well as the continual improvement of the product for improved efficacy and reduced cost of goods sold (COGS). This phase also includes continuing to work closely with legal and regulatory experts as the product is packaged, labeled, and sold and looking for opportunities to extend the product lifecycle and maintain competitive position.

Examples:

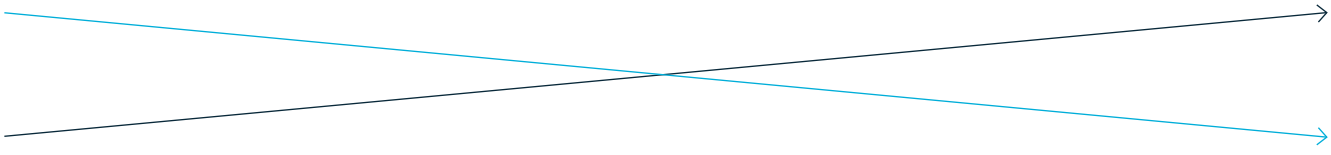
- In the case of crop protection small molecule products, this means conducting large acre field trials in new geographies to demonstrate local product performance and developing new formulations and mixtures with other active ingredients to improve product efficacy, COGS, shelf-life, and meet any local formulation requirements for registration.
- In the case of a digital ag hardware device, it could mean improving usability, durability, battery-life, or accuracy of the product in question.

[12] Good Lab Practices
[13] Good Manufacturing Practices

Phases of Ag Products

Phase 0	Phase I	Phase II	Phase III	Phase IV	Phase V
Define the Problem	**Pre-Field Discovery**	**Early Product Develop-ment**	**Advanced Product Develop-ment**	**Pre-Launch Prepara-tion**	**Launch & Market Expansion**
Establish the size of the opportunity Research the current market solutions Extensively inter-view and meet with farmers and customers	Develop a proof-of-concept solution Prove plausibility under optimal conditions Conduct initial IP review and develop regulatory engagement & IP strategy	Assess safety concerns Establish FTO and execute regulatory engagement & IP strategy Demonstrate performance in more geographies	Generate data needed for any regulatory dossiers Prove scalability and drive down COGS Broaden field testing to even more geographies and test with partners	Finalize regulatory dossier support and stewardship requirements Scale-up production plan Expand market development field trials and develop go-to-market plan	Lower the COGS for production Extend product life-cycle Continue testing to expand into new markets

Risk of products success



Cost and time

not 100% linear

Estimates on Time & Cost of Development

There will be variation across product classes when estimating both the time to market as well as the cost of development. With that acknowledged, this Playbook will use a simple calculation principle for estimating time and cost.

$$\textbf{Time} = \alpha_T + \beta_T$$

$$\textbf{Cost} = (\alpha_c + \beta_c) * (1+\gamma)^{\delta}$$

Where:

α_T = Last reported time for product development
α_c = Last reported cost for developing a product
β_T = Time impact of key research or development trend
β_c = Cost impact of key research or development trend
γ = 2.73 = 10 year inflation rate average
δ = Avg number of years since last reported cost of developing a product

Crop Protection

Disease, Insect, Nematode, and Weed control technologies

Includes seed treatment, in-furrow, or foliar-applied solutions, as well as genetic traits that are used to control pests, pathogens, and weeds to improve crop performance and yield

Crop Protection Product Types

Small molecules
Peptides
Oligos (RNA, DNA)
Proteins
Biologicals
Genetic Traits

U.S. Regulating Agencies: EPA, USDA
Potential U.S. Agencies to be Consulted: FWS, FDA[14]
International testing guideline resources: OECD & FAO

[14] FDA while not required to launch a product, it will be an important agency in the case of any EPA residue studies showing potential impact in food products.

Crop Protection
Product Nomenclature

—

There are many ways to divide up the world of crop protection products. They can be segmented by **application type** (seed treatment, foliar, in furrow, in plant, etc.), **indication** (fungicide, insecticide, herbicide, nematicide, etc.), or **timing of application** (pre-season, mid-season, late-season). For the purposes of the Playbook, we will focus on a segmentation based upon **size**, which also correlates with composition.

The simplified framework below gives a variable range of sizes of each of what we'll categorize as the 4 distinctive groupings:

	Biomolecules		
Small Molecules	**Oligos & Peptides**	**Proteins**	**Biologicals**
			
<1 kDa	5-15 kDa	3-150 kDa	>150 kDa

General Truth:
With an increase in size comes an increase in complexity.

General Misconception:
Because some products have faster paths to market, they will not be as efficacious, specific, or financially viable as the more regulated paths.

Not mutually exclusive:
Final products can contain or be used as a combination of products from more than one group.

Small molecule

Small molecules are a diverse and highly specific group of chemicals that act upon targets (usually proteins) in the cells of organisms. Binding of the small molecule to the target typically leads to the activation or inhibition of that target and its activity in the cell. Small molecules can bind more than one target at a time as well. If we look at PROTACS as an example, these bifunctional molecules bind both the target of interest on one end as well as an E3 ubiquitin ligase on its other end. The number of possible small molecules is enormous and has yet to be fully explored [15]. Small molecules are the most prevalent form of crop protection product used today because they are highly specific to the target(s) of interest, can be synthetically manufactured, and are formulated for shelf stability. The category of small molecules has huge potential for discovering novel products, and we have only scratched the surface of this molecular universe.

Biomolecules

Oligos & Peptides

Oligos and peptides are short chains of nucleic acids or amino acids, respectively, that can be synthesized or produced using recombinant methods. Oligos and peptides find use as the active ingredient in many different types of crop protection products including those utilizing single and double stranded DNA, RNA, small interfering RNA (siRNA), and short amino acid chains. Oligo products in this category that induce RNAi have made their way into the agricultural market in the past decade, and to date there have been at least 18 peptide products commercialized for plant protection including the bioinsecticide Spear®, which originated as a neuropeptide of the venom from the blue mountains funnel-web spider. [5]

Proteins

Proteins are macromolecules consisting of amino acids (and any lipid or carbohydrate post-translational modifications) that can be purified from natural sources or produced using recombinant methods. Proteins can act in a variety of ways as the active ingredient in crop protection products. A well-known example are the proteins from *Bacillus thuringiensis* (Bt), which is a Gram positive, spore-forming species of bacterium from which proteins toxic against a wide range of insects and nematodes have been sourced. These proteins have been successfully used as insecticides against caterpillars, beetles, nematodes, mosquitoes, and flies. Formulations containing Bt cultures have been used in agriculture as pest control applications to the surface of crop plants since the 1950's, and GMO plants expressing recombinant Bt proteins have been widely used for pest control in row crops since the 1990's.

Biologicals

Biologicals are products that contain living microorganisms such as fungi and bacteria as the active ingredient. These microorganisms often have highly regulated metabolic pathways to intake and output key products. A study released in April 2023, conducted by the Stratovation Group and commissioned by the Fertilizer Institute and the Agricultural Retailers Association, found that more than one-third of the U.S. farmers surveyed were currently using at least one biological product on their crops [6]. A University of Nebraska overview of biologicals subcategorizes these products into **biostimulants** like plant growth promoting rhizobacteria (PGPRs), which are used to help plant adapt to abiotic stresses, and **biopesticides** like Regalia®, which is used to control the fungal disease powdery mildew.

[15] For context, using the GBD-17 database compilation of the most frequently occurring atoms with Sulfur (S), Carbon (C), Oxygen (O), and Nitrogen (N), if we combine just 17 of these atoms, it would lead to over 177 billion possible molecules. [4] Going one step further and using 24 possible atomic combinations, we'd easily surpass 10^30 different possible molecules, which is more than the estimated number of stars in the universe. Of course, atoms do not randomly link into molecules, so the actual number of atomic combinations is more limited, but it shows the diversity of small molecule opportunities.

Crop Protection
Small Molecules

Disease, Insect, Nematode, and Weed control technologies.

Seed treatment, in furrow, or foliar applied solutions for control of pest pressure and improvement of crop performance

Crop Protection Product Types

Pheromones
Small molecules
Natural Products

U.S. Regulating Agencies: EPA, USDA
Potential U.S. Agencies to be Consulted: FWS, FDA[16]
International testing guideline resources: OECD & FAO

Estimated overall cost: $312-381m USD[17,18]

[16] FDA while not required to launch a product, it will be an important agency in the case of any EPA residue studies showing potential impact in food products.
[17] Based upon small molecule costs, which have the most well-documented registered products.
[18] The initial costs for the analysis here will be reported numbers from the 2024 public publication from AgBioInvestor on behalf of Crop Life [1] which does a great job of breaking down the trends of crop protection R&D cost over the past 30 years from the top agricultural companies

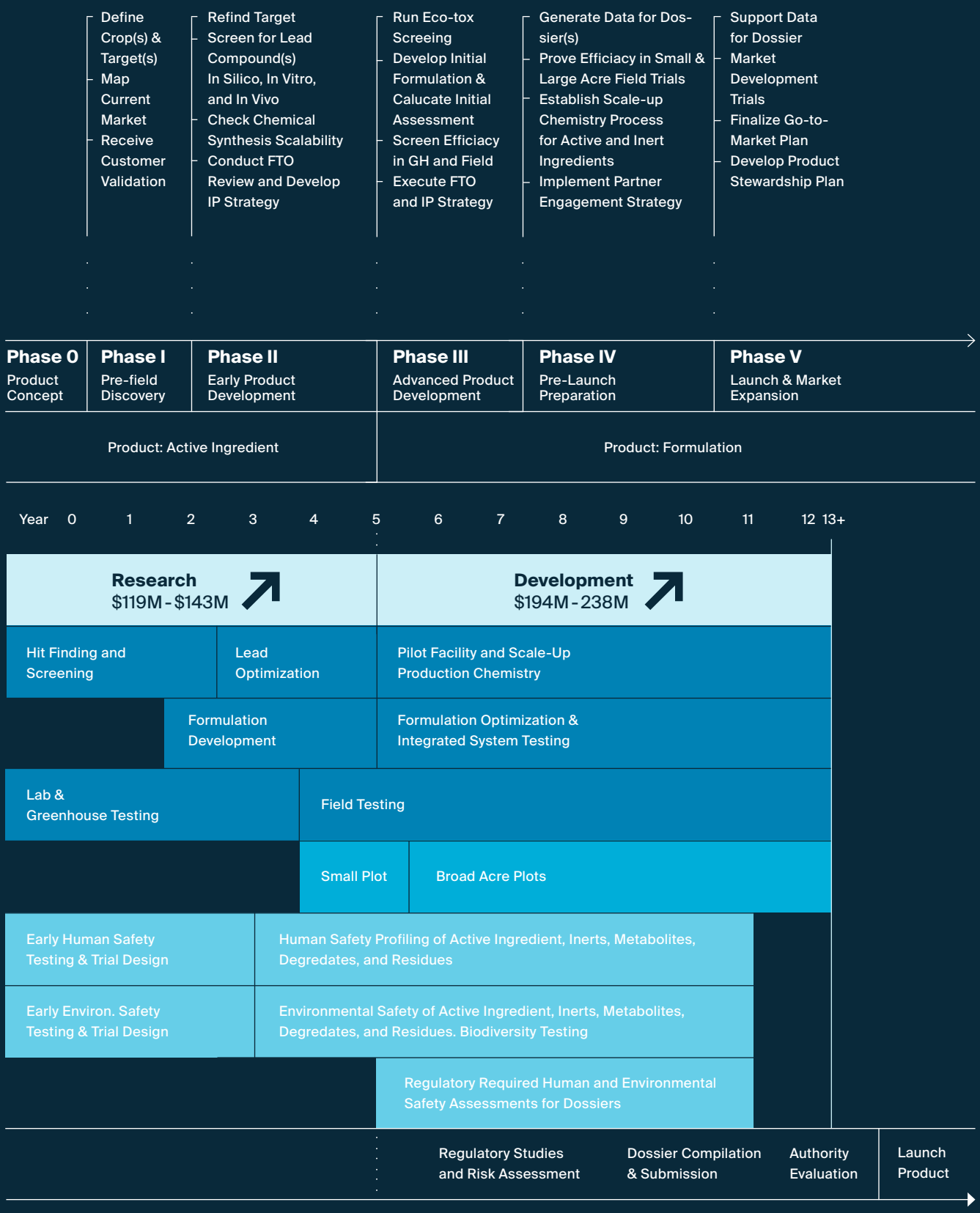
Introduction

Most crop protection products in the market today fall into the category of small molecules. These products are formulated to be used as either seed treatments, in furrow, or as foliar applications to control pests and pathogens including weeds, insects, fungi, bacteria, and nematodes. Crop protection small molecule products represent some of the most impactful AgTech products on the market because of their utility to protect crop yield combined with their specificity, scalable production, and ease of application. The bar to produce these products continues to rise and with it the cost of R&D pipelines. Developing a crop protection small molecule with the highly specific physiochemical properties and safety profile that meets both agronomic needs and societal expectations for safety and environmental protection requires the researcher to consider many factors. Indication, mode of action (MoA), crop segment, efficacy, feasibility of synthesis and production, cost of production, formulation offerings, and safety for humans and the environment are critical attributes for a crop protection small molecule product. The new molecules, like their predecessors, are designed to minimize the potential effects on non-target organisms and the environment according to pre-defined safety and sustainability profiles.

Crop protection small molecule research and discovery can be broken into four basic activities:

Phase 0 – Phase II Research	Phase III – Phase V Development
includes (1) hit finding & screening and (2) lead optimization & formulation development.	includes (3) scale-up of production chemistry and (4) field and registration trials.

Product Pipeline Map (Simplified)



Phase 0 – Phase II: Research

Hit Finding and Screening

Phases 0 & Phase I

Timing

6-24 months

Summary

Target identification is an important part of the early discovery process, and its effectiveness sets the stage for discovering a successful crop protection small molecule. Whether a target has been identified or not, there are two main approaches for discovering small molecules:

Structure-based molecular design: Identifying a specific target to modulate with a small molecule and using computational and combinatorial chemistry techniques to screen small molecules for their potential effect on the target.

Ligand-based discovery: Starting with a small molecule known to have efficacy and using it to dis-cover the target of interest and develop new small molecules with greater efficacy.

The goal is to find novel small molecules (active ingredients) that will effectively modulate a target and cause a phenotypic result.

Opportunities & Common Pitfalls

In Silico ADMET screening allows for better prioritization of scaffolds and chemical series.

Overemphasizing virtual screening without validating both in vitro and in vivo assays. Translating each step from virtual → in vitro → in vivo can be difficult, but important to validate early.

Production costs of any chemistry can become the limiting factor when manufacturing at scale, so finding synthetically facile chemistry improves the chance of making market-feasible product.

Failure to conduct early IP review and develop an IP strategy to ensure freedom-to-operate and patentability of new small molecules.

Cost Range & Trend

$50M-$60M, the highest cost drivers are the establishment of chemical synthesis pathways and the biochemical and biological assays used to rapidly test the efficacy of the small molecule of interest.

Decreasing significantly due to the use of artificial intelligence and computational chemistry tools. These trends mean lower numbers of small molecules must be physically synthesized and tested to find hits.

Please note: These costs are based upon reported numbers from larger ag research companies. Startups that originated their chemistry from academia or focus in a specific chemistry area may minimize much of this early-stage cost.

Target identification is a vital part of the crop protection small molecule research and development process and enables researchers to better understand the **MoA** of potential new active ingredients and optimize the active ingredient for a particular pest or pathogen. Early agrochemical discovery methods relied heavily on phenotypic screens against pests or pathogens of interest. Hits were then studied using extensive genetic, biochemical, and metabolomic methods to determine the molecular target. While this biology first approach still has merit for identifying in vivo hits early in the process, the resulting compounds need to be further screened for undesired activity against non-target organisms and this can lower the hit advancement rate. The target first approach is based on starting with a specific target to modulate in a pest or pathogen and then using computational and combinatorial chemistry techniques to screen small molecules for binding to that target. Target identification is an important part of the early discovery process, and it can enable a higher rate of advancement for crop protection small molecules.

Once a target has been identified, there are several approaches to discover new active ingredients. While there is a wealth of innovation in this space, the most used approaches are structure-based molecular design and ligand-based discovery.

> **Structure-based molecular design** focuses on a game of microscopic Tetris, where researchers attempt to model what molecules might best bind to the target of interest. In most cases the target of interest is the protein's active site or an allosteric site. By binding to these sites, a small molecule can affect the activity of the protein in the pest or pathogen and thus stop it from impacting the crop and reducing yield.
>
> **Ligand-based discovery** leverages existing small molecules known to bind a target of interest and seeks to find similar molecules with improved binding to the same site in the protein or improved chemical properties such as water solubility and ease of synthesis. In this category, researchers need to be mindful of the existing intellectual property rights to ensure any improvements they make are patentable.

Between 1995 and 2014 was the golden age of high throughput screening, where large numbers of compounds were screened to find a single active ingredient that produced the desired response. According to one report, by 2014 developers needed to synthesize and screen over 150,000 compounds to find a single hit [1]. In the last decade, the growth of large, public protein data banks and small molecule databases has enabled focused computational screening, artificial intelligence, and simulation technologies to dramatically decrease the number of compounds that must be physically screened. The use of computational and artificial intelligence tools to mine the large universe of molecular combinations mentioned earlier (over 10^30) has moved much of high-throughput screening from the physical world to the virtual space, so it is quicker and less costly.

Numerous companies specializing in the use of A.I. and computational techniques to accelerate the rate of small molecule discovery have emerged. The utilization of A.I. and neural networks has led to faster and cheaper discovery of potential new hits. Instead of synthesizing and testing tens of thousands of molecules over the span of years, virtual screening has shortened this phase into just weeks and months. Companies can now simulate and rank virtual small molecules and then only synthesize and test a select small number of those molecules. Virtual hit finding has been one of the more compelling uses of A.I. in scientific product development to date. The limiting factor now is the ability to rapidly synthesize and build assays to test the identified potential hits. It is the biology of testing the active ingredient/target interaction that becomes the limiting step in early discovery.

Opportunities & Pitfalls with Hit Finding & Screening

Use of A.I. and simulation in the virtual space has enabled rapid early discovery, however, it further emphasizes the opportunities that still exist in the physical discovery space. The three challenges below highlight recent opportunities in setting up biological assays and producing synthetically facile chemistry.

1	**In silico ADMET screening:**	ADMET (absorption, digestion, metabolization, excretion, and toxicity) screening is important in understanding how an organism will respond to an active ingredient. This is often used in safety assessments. Like hit discovery, ADMET screening models have been migrating to the in silico (virtual) space. In a recent publication, researchers leveraged a deep learning approach to make predictions of 100 ADMET assays, assessing the potential for a compound to become a relevant drug candidate [7]. However, the species of interest in agriculture do not follow the same ADMET rules as those used for humans. This was supported in 2001 by Colin Tice, when he published his results when looking at Lipinski's rule of 5 as it applied to agrochemicals [8]. In silico generated virtual compounds generated through both optimized binding models and ADMET models can allow for better prioritization of scaffolds and chemical series, but the screening parameters will need to be adjusted.
2	**Translating from in silico to in vitro to in vivo**	Translating from in silico to in vitro to in vivo can be difficult but is important to validate early. Testing both in vitro (for example in a 96 well plate or test tube) and in vivo (in a model organism or pest) can be simple and inexpensive in agriculture, but it takes time to design and execute the right assay.
3	**Finding an economically viable chemical synthesis route:**	Chemical synthesis can become the limiting factor when manufacturing at scale, so finding synthetically facile chemistry improves the chance of making a market-feasible product. The chemical synthesis route taken to make the active ingredient for assays and screening in this phase is rarely the final process used for production, but ensuring the active ingredient can be made or bought at a reasonable cost and in sufficient quantities early on improves the likelihood of success.
4	**Conducting an early FTO Review and developing the IP protection strategy**	Ensuring both freedom-to-operate (FTO) and the ability to protect future products will allow for value capture on products and eventual return on investment.

Hit finding and screening has become one of the most technology-transformed research steps in agricultural discovery due to the use of artificial intelligence and automation. This also demonstrates the potential of these technologies to optimize other stages of R&D testing pipelines to become cheaper and faster while remaining equally safe.

EXAMPLE

In silico

An increase in the number of molecules and protein structures made available in public databases has enabled massive virtual screening and prioritization of potential small molecule hits that may modulate the target protein and have a preferred ADMET profile.

In vitro

A target protein of interest can be tested in a 96 well plate with a selection of small molecules by leveraging high-throughput techniques to screen for binding [26]. The researcher can use these results to make a much stronger correlation to the MoA for an active ingredient

In vivo

Rather than just limiting the assay to the target protein, the researcher can put an entire insect or pathogen of interest in the multi-well plate and expose it to varying concentrations of the active ingredient while measuring the impact in a dose- and time-dependent manner. This will allow the researcher to evaluate the **efficacy** of an active ingredient.

Lead Optimization
& Formulation Development

Phase I & Phase II

Timing

12-36 months

Summary

Lead optimization and formulation development aims to take the understanding of the physical and chemical properties of an active ingredient and develop a seed treatment, in furrow, or foliar spray system.

Lead Optimization helps answer key questions to improve the efficacy and safety of the active ingredient:

1. How is the active ingredient absorbed and how will it move in the species of interest?

2. How strongly does it bind to the target of interest and how quickly does it generate a response?

Formulation science requires analyzing and developing how an active ingredient can be used with other active ingredients and inert ingredients, provide consistent binding to the target, minimize the use of safety limiting ingredients, and have a path to being produced profitably at the scale needed.

Human and environmental safety studies are vital to ensuring that a formulated product has the desired impact on the target organism with minimal off-target effects.

At the end of formulation development, the product is the formulation and not just the active ingredient.

Opportunities & Common Pitfalls

Evolving regulatory requirements. Regulatory requirements on micro-plastics and other formulary ingredients are evergreen. Product developers must seek paths for their products to work with biodegradable and environmentally minded ingredients.

The mixture challenge. Novel active ingredients need to be tested in combination with other products that will be applied at the same time (tank-mixed or sequential) or using the same equipment.

Volumes of application. With the emergence of precision application, farmers are demanding formulations be built with application volumes at the forefront. This is paramount as spray systems move toward low, very low, and ultra-low volume formulations.

Initial COGS assessment. For the ingredient(s) included, it is important to calculate early on if they will all scale or if there are limits on material supply.

Cost Range & Trend

Costs can range between **$68-83M**

Increasing with the requirements for toxicology and environmental testing.

Once the number of active ingredients for the lead molecule have been identified in early screening, they need to be optimized for performance. **Lead Optimization** & **Formulation Science** seeks to achieve three product characteristics: **Efficacy, Safety, and Manufacturability.**

Lead Optimization

Efficacy becomes the word of choice, when considering how well a product will work on its target of interest. The point of pursuing efficacy is to get the maximum response with the minimum amount of active ingredient. This phase also is aimed at answering the question: "Does the active ingredient consistently do what it needs to do?" The research team already started answering this question in the discovery phase and can now further optimize the active ingredient performance. While many measurements are used to define the quantitative structure-activity relationship between a target and active ingredient, the metrics below are foundational to answer optimization questions.

How is it absorbed and how will it move in a species of interest?	**pKa –** Acid-based dissociation constant that defines the acidic strength of a molecule in a solvent. The lower the value the more acidic the molecule. If a molecule has limited solubility in water, researchers will often determine pKa values in other solvent mixtures such as water/dioxane in which the molecule may be more soluble.
	LogP – Partition coefficient provides indications on whether a substance will be absorbed by living organisms or be easily carried away and disseminated by water. It measures the ratio of the organic solubility to the water solubility [9] and is usually calculated with an HPLC or shake flask methodology.
	LogD – Distribution coefficient that is a pH-dependent version of LogP and used to measure lipophilicity.
How strongly does it bind to the target and how quickly does it generate response?	**EC50 –** Concentration of the active ingredient that produces 50% of the maximum response from the target. There is no hard and fast rule as to what concentration is good enough to continue lead optimization, but usually researchers will want to see activity of at least at 1µM or less to move forward, with optimum activity hopefully occurring more in the nanomolar range.
	IC50 – Similar to EC50, this measures the concentration of a given inhibitor where the binding to the molecular target reduced by 50%.
	LC50 – Same as the EC50 but the phenotype measured is death of an organism.

Dosing studies are meant to be conducted both *in vitro* and *in vivo*. These studies require sourcing biological material to test, and the readout results should be simple and direct measurements of efficacy. The results of these assays also serve as a control benchmark for future testing of the active ingredient and as a baseline for comparison in future formulation testing under specific delivery mechanics.

Generating *in vivo* and *in vitro* proof of concept is generally advisable prior to considering moving forward into formulation optimization. Basic protein binding assays *in vitro* are key for hit validation, but doing additional whole cell or whole species assays will better capture multi-variate factors of an organism. While it might seem complex, it is often simpler to set up a cell-based assay or whole species assay to get clear results demonstrating whether the product is efficacious against the weed, insect, or pathogen of interest. These results also can be used with genetic analysis to better understand if there are potential mutations that, if present in the target of interest, will minimize the efficacy or durability of a new hit molecule in the future.

Formulation Sciences

Perhaps the most complex and opaque piece of the crop protection small molecule discovery is formulation development. There are very few university programs with a specific emphasis in formulation chemistry. Instead, training in this field is approached as a multidisciplinary study that combines biochemistry, physical chemistry, organic chemistry, and molecular biology principles. There are many different types of formulations used in commercial agriculture, but here we will focus on seed treatment formulations and foliar spray formulations.

Seed Treatment & Foliar Formulations
The most common type of formulations in the market fall into one of the six categories [10] below.

Type of Formulation	Abbv.	Description	Examples
Suspension concentrates	**SC**	Water based solution that requires having a melting point higher than room temperature. Seed treatments are often a version of suspension concentrate that have supplemental additives for nutrition and adhesion to the seed.	Fluopyram, Imidacloprid
Emulsifiable concentrates	**EC**	Usually liquid in a dispersed oil phase in an aqueous solution, combines the active ingredient with aromatic solvents that have a lower cost of manufacturing and are easily polarized. Emulsion droplets aim to be between 0.1 and 1.0µm	Prothioconazole
Capsule Suspension	**CS**	The active ingredient is encapsulated in a small microcapsule made of a polymer shell and mixed with a suspending agent. There is movement away from polymer shells to more biodegradable solutions.	Acetochlor
Soluble Liquid	**SL**	Simple water-based solutions that are used where the active has a high solubility. SLs scale easily and make up a lot of the market today.	Glyphosate, Glufosinate
Water Dispersible Granules	**WG**	Conveniently packages a solid granule that when mixed in a tank will dissolve in water into a fine particle suspension. [11] Often requires a wetting agent to help with better dispersal.	Atrazine
Wettable Powders	**WP**	Dry formulation that is leveraged for active ingredients that have high melting points and water-insoluble solids. WPs are mixed with water and other inerts, diluents, and surfactants prior to spraying.	Cypermethrin

Choosing which type of formulation to pursue begins with consideration of the crop and geography, and then moves to considering the properties of the active ingredient. As an example, it is unlikely a water-soluble liquid formulation will work for a highly hydrophobic active ingredient unless other technologies are used to provide the ability to mix soluble and insoluble actives together.

In addition to the active ingredient, a formulation also includes other ingredients that each have their own function and safety profile. These are usually known as the inert or non-active ingredients. Do not let the name inert fool you – these ingredients each have their own impact in the formulation. The regulatory profile of these components is significant and can vary by geography. The global and regional acceptance of inert ingredients should be assessed early in the formulation development process as failure to do so could be problematic in later stages. Some of the main classes of inert formulation ingredients are summarized here:

Surfactants (i.e. wetting agents)	Constructed of both hydrophilic and hydrophobic parts to enable oil and water to mix. A wetting agent is a type of surfactant that reduces the surface tension between two substances. A surfactant can come with an electrical charge (anionic or cationic) or not.
Diluents	The main solution that the active ingredient is diluted into. In the case of an SL formulation, it would be water. In the case of an EC, it might be petroleum-based solvent.
Fillers	Bulking agents added to the formulation for mass.
Binders	Leveraged in WG and WP formulations to enable resistance to physical and chemical stresses to reduce dusting.
Dispersing Agents	Works into the diluent to ensure that the active ingredient does not amass together in the solution and stays evenly mixed and spread when applied.
Encapsulates	Polymers or biologically-derived encased technology that isolates the active ingredient from the water or oil based formulation.

Formulations combine active and inert ingredients to yield a safe, application-compatible, stable product. While unique in their physical and chemical properties, each inert ingredient either ensures that the active ingredient effectively reaches its target of interest or supports the formulation stability. **At the end of Phase II, the product of the RESEARCH phase changes from being the active ingredient to the entire formulation**, and **the efficacy and safety of the entire formulation will be submitted for review by regulating authorities.**

Safety

Crop protection small molecules and their formulations aim to have the desired impact on the target organism with little to no off-target effects. In cases where there is potential for exposure to derivatives or intermediates of the active or inert ingredients, these too will have to be studied.

Human and Animal Safety and risk assessments include evaluation of the product in numerous safety studies, such as: (1) toxicological studies to determine potential adverse effects on all major organs (liver, brain, thyroid, reproductive organs, nervous system, etc.); (2) carcinogenicity, mutagenicity, reproductive toxicity, and endocrine disruption studies; (3) absorption, distribution, metabolism, and excretion (ADME) pathway studies; and (4) determination of the maximum dose at which No Adverse Effects (NOAEL) are observed in test species. Please note that NOAEL is a U.S. federal standard and may not comply with individual U.S. state or foreign agency requirements. Requirements for each type of study should be reviewed at national, state, and local levels for the desired market on a country-by-country basis. For products that will be used on crops that serve as animal feed, such as maize for making silage, a risk assessment of livestock animals will be necessary. Additionally, the list of animals for safety testing has continued to evolve and can require additional testing and proof of safety across a broader species panel.

Environmental Safety requires experimental studies and modelling, which authorities evaluate during the product registration process. In regions with well-developed programs to regulate crop protection products, safety assessments are based on the specific regional or national safety standards. When looking to register a product for use in a country with less developed standards, a common approach is to meet the safety standards of relevant regulatory regions and countries and assess if the product can be safely used under local conditions and existing regulations.

Opportunities and Pitfalls with Formulation Sciences

Formulation science deals with multiple challenges from extending shelf-life stability to improving the dispersion and delivery of an active ingredient. The challenges below highlight some of the recent opportunities created by the emergence of both technology and new policies around the world.

1	**Evolving regulatory requirements**	New requirements for environmental safety can emerge based upon continuous policy changes. As an example, the EU has recently passed Commission Regulation (EU) 2023/2055 restricting synthetic polymer microparticles. This will lead to the need for alternatives to many of the microplastics used in capsule suspensions.
2	**Mixture Challenge**	The use of a single active ingredient repeatedly can lead to resistance developing in the target. Therefore, it is important to work on formulations that enable the use of multiple active ingredients with different modes of action in combination to decrease the development of resistance. Each active ingredient will have unique sensitivities to moisture, pH, or temperature and may interact synergistically or antagonistically with other active ingredients.

The use of encapsulation and oil dispersion are enabling more active ingredients to be safely used together, and this area of research continues to grow. For a startup developing a new active ingredient, it can be both costly and time consuming to make a mixed formulation for use with other active ingredients. Some products may be sold as stand-alone products, but for any product that will be used in combination with others the first step should be ensuring tank mix compatibility with a secondary focus on mixture products.

3 **Delivery & Low Volume Formulations**

Ground-based and precision drone applications of crop protection products in agriculture are enabling the use of even lower dosing requirements. This can lead to lower costs for the farmer and reduce the risk of off target environmental impacts. With the rise of precision application came some new terms that do not yet have a consistent market definition. While some definitions are being developed [12], [13], [14], [15], for simplicity's sake in this document we will use the following definitions:

Type	Volume Application Rate
Standard Formulation	= or >50 L/Hectare
Low-Volume Formulation (LV)	49-20 L/Hectare
Very-Low Volume Formulation (VLV)	20-10 L/Hectare
Ultra-Low Volume Formulation (ULV)	<10 L/Hectare

ULV formulations have led to the use of ingredients that remain fluid and spread to improve coverage or change bioavailability through localized concentration gradients to have inelastic efficacy and performance at lower application volumes. Additionally, the goal of these formulations is to achieve better stickiness to vegetation as well as lower drift risk.

4 **Conduct an initial COGS assessment**

Once the product, including all active and inert ingredients, is better understood it is important to calculate if it will scale or if there are limits on material supply. Ingredients that are dependent on rare supply may either have cost or supply chain limitations that prevent the formulation from becoming economically viable at scale. This assessment of a formulation's manufacturability is revisited frequently as the chemical synthesis and chemical process approaches are continuously improved (see next section).

Cost of Research

—

From 1995 to 2019 the cost of hit discovery, lead optimization, and formulation development that occurs between Phase 0 and Phase II increased from $72M to $127m in large part due to the growing cost of chemistry discovery and biological screening [1]. Today, the use of in silico computational techniques has reduced the upstream cost associated with the discovery of lead compounds. Now, fewer compounds must be chemically synthesized for biological screening in vitro and in vivo to discover and optimize a new active ingredient (hit). Unfortunately, although virtual screening was used frequently in the 2000's and lead to significant reduction in costs for hit discovery, the savings have not yet been reflected in the reported costs for new product development.

Key Trend 1

A more efficient in silico program should lead to a smaller number of potential hits that require biological assays to screen for an efficacious compound. This should enable a faster path into Phase II Lead Optimization. While some discovery programs claim more dramatic numbers, one assumption is that at least 25% of the chemistry costs from the 2014-2019 surveyed costs of $64M have been mitigated down to $48M, resulting in a **$16M decrease**.

Key Trend 2

Early toxicology and environmental testing costs are expected to continue to rise as extra experiments, time, and resources are needed to meet the increasing safety testing requirements. Costs for safety testing grew from $7M to $11M between 2010 and 2019 (~50%). Using a consistent growth rate between 2019 and 2024 (5 years) would have had costs grow another 25% or a **$2.75M increase**.

$$Cost = [(Last\ Reported\ Cost) \pm Key\ Trends] * 10\ year\ inflation\ avg^{5\ years}$$

$$\$\mathbf{130}\boldsymbol{m} = [\$127m - \$16m + \$2.75m] * (1 + 2.73\%)^5$$
$$\text{(Hit Disc)}\quad \text{(Eco/Tox)}$$

[1], [16]

This report will refrain from providing an exact number but uses a 10% variance range to estimate that the **cost of Phase 0 – Phase II Research is between $118M and $143M.**

Phase III – Phase V:
Development

Scale-up
Production Chemistry

Phase III & Phase IV

Timing

36-48 months

Summary

Scale-up production chemistry focuses on finding an economical, safe, and scalable chemical synthesis route that is process optimized for producing the active and any other key ingredients or their pre-cursors in a formulation.

Once a chemical synthesis route is found, companies must make the decision to either externally source, contract/toll manufacture, or internally build capacity to produce the final ingredient(s) for a formulated product. The production method decision hinges on a few key considerations including **economics**, **safety**, **logistics**, and **policy**.

Coming out of this area of research, a company should have a process chemistry pathway defined for the active ingredient, a pilot production facility built or made available, and production strategy for any key ingredients and any rate-limiting pre-cursors that is de-risked from global and commodity market considerations.

Opportunities & Common Pitfalls

Diversify the production supply of key raw materials, pre-cursors, reagents, and ingredients(s) protects from future supply risks.

Sourcing more sustainable starting materials without sacrificing on COGS.

Reaction byproducts and heat must be managed to minimize scale-up production risk and enable the recycling of energy and by-products.

Trust but verify. Staying on site for installations can help avoid issues and provide better collaborate with builders in case any issue or need to pivot arises.

Cost Range & Trend

Costs can range between **$31-38M**

Maintaining flat due to the ability to access pre-existing infrastructure while there is high susceptibility to the cost of raw materials.

This chapter will not specifically get into the science of **chemical synthesis** and **chemical process optimization** foundational for scaling up the production of a new product, but brief overviews are provided below:

Chemical synthesis refers to the culmination of all the reaction steps taken to get from a hit molecule to the final active ingredient. There can be a single or multiple chemical reactions needed to produce the final molecular product. Each one of these steps is a reaction that requires its own optimization. An overview of chemical reaction optimization can be found in the 2023 A Brief Introduction to chemical reaction optimization [17].

Chemical process optimization is the system engineering methodology used to improve the efficiency and profitability of a chemical process by implementing manufacturing procedures that improve the yield of desired product, minimize waste production, reduce energy consumption, and improve process safety. An overview of this can be found the 2022 editorial Integration and optimization in chemical process industry [18].

Whether the final product is internally manufactured or outsourced to a toll manufacturer, building or having access to a pilot facility can be critically informative for chemical process optimization. Pilot facilities allow for the continuous optimization of the synthetic and mechanistic processes involved in chemical synthesis. Pilot facilities enable quick process chemistry support and are key for early-stage synthesis of the product candidate for testing.

As the active ingredient moves through development, researchers often find that the initial chemical process is not the best option for larger-scale production. For example, the initial synthesis route might require expensive reagents, present challenging operating conditions, or create possible safety hazards when performed at larger volumes [19]. In addition, reaction times and complicated product isolations may lead to lowered capacity (throughput), higher equipment demands, and higher production costs. Stepwise evaluations of the chemical process allow for the collection of process data and a fundamental understanding of the opportunities for optimization. Building a pilot plant supports the planning and management of secondary reaction products, management of heat and carbon emissions, and informs understanding of the reaction chemistry efficiency at scale.

The decision on whether to build manufacturing capacity or access toll manufacturers includes considering economics, safety, logistics, and policy. Key questions are:

Economics	How can the product most cheaply be produced via the optimized chemical process, factoring in both capital facility and continuous operational costs?
Safety & Logistics	How can the product and any of its pre-cursors safely be made and reliably delivered to its destination with minimal risk to employees and the environment?
Policy	Is the chemistry production and manufacturing of the final product or any of its pre-cursors impacted by any regional, national, or international policy?

These considerations in combination will help inform the best path of production. As an example, a startup may consider toll manufacturing with an international contract manufacturing organization (CMO) who operates at a lower cost. However, when logistical shipping concerns, product stewardship, and international policy between the startup's country and the CMO location country are considered, the decision may change.

Opportunities and Pitfalls with Scale-up Production Chemistry

1	**Diversify production supply**	A startup may also consider minimizing its production supply risk by having multiple manufacturing relationships. Having a diversified supply chain decreases risks associated with the logistic and policy considerations mentioned previously. Examples have arisen where global trade has slowed down due to limitations on waterway throughput or weather pattern changes. Policy risk includes taxation, currency exchange, and stewardship as considerations that make the case to ensure supply from more than a single provider for any key ingredient, precursor, or intermediates.
2	**Sustainably sourcing starting materials**	The decision on where to source starting materials is an important consideration. If a CMO can produce most of your starting material at lower cost, it can be simpler and cheaper to source from them rather than build manufacturing capability. Supply chains are often spread across multiple manufacturing sites, so isolation of intermediates and shipping to other facilities for downstream reactions will require registration and tracking. Today, the full life-cycle-analysis of any product's carbon impact is a growing consideration, but working with multiple manufacturing sites to optimize cost and diversify risk to the product supply can possibly increase the carbon footprint when factoring in shipping between sites. Finding suppliers who sustainably produce starting materials, intermediates, or the final product while minimizing the distance they travel can lead to reduced emissions.
3	**Reaction by-products and heat must be managed**	Exothermic reactions that produce heat may not be an issue at lab or pilot scale but could become an issue when metric tons of material are being produced each day. Closed loop facilities that leverage their own exothermic reactions as energy for the endothermic reactions have a better carbon footprint. Likewise, many secondary products considered waste by some can be leveraged as inputs for creation of new additional products. As an example, methanol is a byproduct of many reactions and may be recovered for use as a solvent or reagent in another process. The recovery and use of byproducts are very common practices utilized by integrated chemical manufacturers to reduce costs and waste.
4	**Trust but Verify**	Too often when installations are contracted out the 3rd party company is trusted to do the appropriate installation. When thinking about the testing and scale for a startup, time is critical. Having the innovator's own process chemists and engineers on site to collaborate on all installations is critical to ensuring success. Failure to do so may lead to lack of oversight that can cost months of testing time. Even if an installation delay has not increased a company's construction costs due to insurance/guarantee of installer, the time lost can translate to a season of product for the agricultural market. Real-time analytics on the reaction process and composition also can lead to quicker optimization of the chemistry.

Scale-up production requires the expertise of chemists and engineers to come together to manufacture innovation at scale and drive down COGS of new products.

Field and
Registration Trials

Phase II, Phase III, Phase IV, and Phase V

Timing

72-84 months

Summary

Conducting field and registration trials is the most expensive activity in bringing a new crop protection product to market. This makes it even more critical to ensure special attention is paid to experiment design, site selection, mixing products, and stewardship and regulatory requirements.

Experiment Design and **Site Selection** can minimize weather and random effects, allow for measurement of the null hypothesis, and ensure statistical power of field trial results. Testing partnerships serve as early market and business development to achieve belief among the future customer base.

Mixing products answer the question on whether a given crop protection product has a synergistic, antagonistic, or neutral effect when combined with other products that would be applied to the field at the same time.

Following the **regulatory requirements** set by the appropriate agencies and **stewardship best practices** will ensure that all testing, including residue and decline trials, are well executed and drive a quick and safe path towards product registration.

Coming out Phase IV the product should be fully registered and efficacy well defined. Phase V going forward will focus on broadening field testing and data generated to support market expansion opportunities.

Opportunities & Common Pitfalls

Engage farmers and support organizations. By engaging early, researchers can ensure support, advocacy, and 3rd party verification by market partners.

Missing Positive Controls. Failing to test early enough with the appropriate positive control(s) may mislead the researcher about the efficacy of the product. Crop protection should be testing potential new products in the full system of products that a farmer is using.

Underestimating Field Trial Costs. Contracted growers often expect a premium over commercial operations and generating additional data points may come at additional cost.

Cost Range & Trend

Costs can range between **$163-200M**

Increasing significantly due to the cost associated with conducting the necessary field trials to:

1) Meet and satisfy the regulating authorities.
2) Meet and satisfy the testing demands of customers and distribution partners.

Field trials are the costliest effort in most agricultural research pipelines. There are specific and strict requirements when it comes to registration trials, but most fields trials aim to answer the question: Does this solution safely work in the field with the current and future agronomic practices across a variety of geographies, soil, and weather conditions?

While the question seems simple enough, if you break it out into its components there are multiple questions that each require their own experimental design.

- Crop safety and environmental persistence studies across multiple environmental conditions
- Impact of performance of product by farm practice (tilling, irrigation, etc.)
- Impact on soil microbiome and nutrient composition
- Weather pattern correlation studies (days after rain)

Many of these studies can be combined for greater power analysis, however, others may not apply. Remember the more data collected on each trial provides better product understanding, with the tradeoff of higher cost of data collection. Researchers can opt to take soil samples at every field, however, to do so can increase the cost of a field trial based on labor/effort.

When looking at establishing a field trial testing program for a crop protection product, key areas to focus on are:

1 **Experimental Design**
2 **Site Selection**
3 **Mixing products**
4 **Stewardship or Regulatory requirements**

1 Experimental Design

Experimental design is critical to answer key questions about how a crop protection product will work in the field. The design of an experiment should include all the products of interest to be tested as well as proper control groups. This should include a basic untreated control and a positive control group. The number of plots included in an experiment, number of replications, and number of fields and locations where an experiment (trial) is run will determine whether a researcher can prove with statistical significance that their product works. In the registration section below are links to EPA, USDA, and FDA guidance on **registration trials** with specific requirements on this. When researching product efficacy, four of the most frequently used experimental designs are illustrated and summarized below.

Random Control Block (RCB)

RCB experimental design randomizes which plot gets what type of treatment (entry). If Entry 1 was low treatment, Entry 2 was medium, and Entry 3 was high treatment, then by randomizing where they appear in single replication of an experiment the researcher can overcome any field-specific bias. What kind of biases are being avoided? For example, picture all the Entry 3s are kept in the right most column of the example here. This column might be the closest to a road or river. Dust from the road or the potential flood of the river might impact the performance of that right most column of plots differently than the other columns. Were that to happen, all data on how Entry 3 performed in a field would be lost. In an RCB design with 4 replications (as visualized), the design

ensures that only one plot worth of data with regards to Entry 3 is lost. The use of border plots or other control factors can also mitigate environmental effects, but an RCB design allows for mitigation of both environmental effects and human selection bias.

Randomized Control Block

Control	Entry 2	Entry 3	Entry 1
Entry 2	Control	Entry 1	Entry 3
Entry 1	Entry 3	Control	Entry 2
Entry 3	Entry 1	Control	Entry 2

Replication 1
Replication 2
Replication 3
Replication 4

Group Block Design (GBD)

GBD is like RCB design except not every entry is randomized in the experimental replication, rather the entries are grouped by a shared attribute. In the example here, entries are grouped by which adjuvant was added to the formulation and within each group the entries are then randomized. Groupings can be of either equal or unequal sizes. It is important to remember the entries within a group remain together but are placed in random order within that group. The groupings then themselves should be randomly assigned.

Group Block Design

Entry 6	Entry 6	Entry 6	Entry 1	Entry 4
Entry 9	Entry 10	Entry 7	Entry 8	Entry 5
Entry 15	Entry 13	Entry 17	Entry 18	Entry 16
Entry 12	Entry 11	Entry 14	Entry 20	Entry 19

Group 1	Adjuvant 1
Group 2	Adjuvant 2
Group 3	No Adjuvant
Group 4	Adjuvant 3

Split Plot & Strip Plot

Split plot and strip plot trial types are variations on the GBD where within large plots two variables are tested simultaneously. Variable A would be tested in vertical strips and Variable B would be tested in horizontal strips. As an example, Variable A could be a crop protection small molecule product and Variable B the irrigation volume. This generates a gradient of performance and interaction effects for the researcher analyze.

Side-by-Side

Side-by-side trials are a variation on the GBD where multiple replications of a two-entry test are leveraged. These are usually conducted with large scale plots and are often a best-in-class methodology to test a late-stage product against the positive controls currently used in the market. Side-by-side replicated trials are often used in the development of marketing material and commercial sales numbers.

2 Site/Location Selection

The goal of fields trials should be to remove as many variables as possible, including weather, to best assess the direct impact of the product. Early in the product testing cycle field trials will be conducted at a smaller number of locations to test general efficacy, and then later in the product testing cycle a larger number of locations will be tested enveloping a wider range of environmental variables. When setting up field trials for crop protection, it is critical to accurately measure the presence of the pest of interest that the product is looking to address. Leveraging historical data and predictive tools, researchers can better select fields where there is likely to be sufficient pest pressure. When establishing trial design in these areas, the experimental design will allow more immediate comparisons to the performance of fields with and without the product, thus allowing for better testing of the **null hypothesis**. The null hypothesis is the comparison of the product to how a crop would perform in the absence of any treatment. In the case where there is pest pressure, the researcher would expect to see a negative impact from the pest on the plots with no treatment. However, if there is not sufficient pest pressure at the location, then the untreated acres may look exactly like the treated acres. This emphasizes the importance of location selection for pest pressure when setting up trials. Certain testing sites and locations can also be approved for inoculation studies, where the pest of interest is intentionally introduced to test products. These sites are highly regulated, and researcher will need to work closely with the appropriate agencies in these trials.

3 Mixing Products

If the basic control group will have no crop protection product applied and the positive control group will have modern best practices applied, then it is important to run a mixed product analysis. This will answer whether a crop protection product has a synergistic, antagonistic, or neutral effect with the other products applied to the field.

In the example of a new seed treatment for fungicidal control, seed treatments can be formulated and layered to minimize interaction between products on the seed. However, there is only so much that can be added to the seed before it affects the seed flow rate through a planter.

Also, as the coating of the seed breaks down, the layers of treatments may begin to interact with each other. If a new fungicidal seed treatment and biological seed treatment for seed germination are layered, the researcher will want to know how they interact. If the fungicide has a negative effect on the seed germination treatment, it is unlikely that these products will be stacked and one would have to be sacrificed or applied in a different manner.

In foliar applications, crop protection products are often tank mixed or sprayed at the same time. Testing which products can be tanked mixed is usually done during formulation testing, and this will continue in later stages as multiple products are spray applied in the field. It is important to analyze by crop and growing stage what the most frequently used products are and to establish trials that integrate the new product into current field-testing spray and irrigation practices.

4 Stewardship or Regulatory Requirements

Crop protection products are one of the most stringently regulated products in agriculture today. Before crop protection products can enter the market and be used by farmers, they undergo country-dependent evaluation and approval by local authorities for each targeted country to ensure that they may be used safely under local conditions [20]. If the product is entering the international commodity market, the product developer (or importer depending on the agreement) should seek not only product authorizations in the country where the product will be manufactured and sold but also import tolerances/authorizations for key import countries to follow accepted industry standards. The International FAO Code of Conduct for plant protection products and the regulatory standards of Organization for Economic Co-operation and Development (OECD) set minimum standards for testing. Import approvals require time and focus to work with the appropriate agencies. Scoping the scale of your product reach early on will allow for planning how to best engage regulating authorities globally.

In the U.S. as an example, crop protection products are regulated at the federal, state, county, and local levels. Compliance at the federal level includes[19]:

- **F**ederal **I**nsecticide, **F**ungicide, and **R**odenticide **A**ct (**FIFRA**) enforced by the EPA ensures that products do not cause unreasonable adverse effects on human health or the environment.
- The **F**ederal **F**ood, **D**rug, and **C**osmetic **A**ct (**FFDCA**) enforced by the Food & Drug Administration (FDA) requires the establishment of legal limits for pesticide residue in or on agricultural commodities.
- The **E**ndangered **S**pecies **A**ct (**ESA**) administered by the **F**ish and **W**ildlife **S**ervice (**FWS**) and National Marine Fisheries Service (**NMFS**) ensures that any action authorized by a federal agency will not likely jeopardize the continued existence of any listed species and is managed by services outside of the EPA.

In the United States, state pesticide laws govern the use of pesticides at state level. All federally approved pesticides must also be approved at the state level.

[19] There are also transportation, occupational, and advertising regulations to consider

When conducting R&D field trials, **stewardship is important**. Below are seven key principles[20] of stewardship that should be applied during crop protection small molecule product development.

1 Obtain country-specific important approvals and testing permits or authorizations for non-registered products.

2 Experiments/trials including the use of experimental products will be conducted by trained personnel who wear appropriate personal protective equipment (PPE) as determined by the human risk assessment.

3 Protocols and procedures are established when performing trials to prevent non-registered products from entering the food or feed chain.

4 Experimental products, seeds, and plant materials will be labeled clearly, meet applicable regulatory requirements and include information about safe handling.

5 Reasonable efforts will be made to ensure limits on potential cross contamination of any experimental product.

6 Crops and harvests from trials with non-registered products will be destroyed, unless otherwise allowed by regulations and laws.

7 **Residue trials for crop protection products will be conducted in accordance with national/regional regulatory requirements prior to marketing** such as in accordance with Codex Alimentarius and FAO guidelines.

Opportunities and Pitfalls with Field and Registration Trials

1	**Engage Farmers and support organizations**	Growers associations, farm bureaus, research farms, and land grant universities all help advance and bring the best possible products to market to improve farm productivity. Engaging early in the testing process of a new product with these organizations empowers them to provide their own resources to ensure successful trialing of new products and advocacy of the results generated. There are over 100 organizations in the U.S. alone dedicated to improving farm productivity and farmer access to resources.[21]
2	**Missing Positive Controls**	Too often, early-stage products are only tested against the null hypothesis, but it benefits researchers to ensure that proper positive controls are included in early-stage experiments. A common positive control is the current form of best practice for controlling

[20] Not an all-inclusive list

[21] The following Wikipedia page gives a guide to some of these organizations, however, please note that this list is NOT all inclusive.

Control	Control	Control	Control	Control	Control	Control	Control
Control	Entry 1	Entry 1	Entry 1	Entry 1	Entry 1	Entry 1	Control
Control	Entry 1	Entry 1	Entry 1	Entry 1	Entry 1	Entry 1	Control
Control	Control	Control	Control	Control	Control	Control	Control
Control	P.Control	P.Control	P.Control	P.Control	P.Control	P.Control	Control
Control	P.Control	P.Control	P.Control	P.Control	P.Control	P.Control	Control
Control	Control	Control	Control	Control	Control	Control	Control
Control	Entry 2	Entry 2	Entry 2	Entry 2	Entry 2	Entry 2	Control
Control	Entry 2	Entry 2	Entry 2	Entry 2	Entry 2	Entry 2	Control
Control	Control	Control	Control	Control	Control	Control	Control
Control	Entry 3	Entry 3	Entry 3	Entry 3	Entry 3	Entry 3	Control
Control	Entry 3	Entry 3	Entry 3	Entry 3	Entry 3	Entry 3	Control
Control	Control	Control	Control	Control	Control	Control	Control
Control	P.Control	P.Control	P.Control	P.Control	P.Control	P.Control	Control
Control	P.Control	P.Control	P.Control	P.Control	P.Control	P.Control	Control
Control	Control	Control	Control	Control	Control	Control	Control

Entry 1	Low volume
Entry 2	Mid volume
Entry 3	High volume
Control	No treatment
P.Control	Standard Practice

a pest of interest. This can be anything from an existing crop protection product to simply ensuring a crop rotation cycle to avoid persistence of a pest. For any new product to succeed in the market, it must demonstrate a significant performance and/or cost advantage over the existing practices and products in market. Therefore, it is critical to include a positive control in the experimental design. The example design below highlights how an experimental trial might be constructed for a given field.This design shows one replication of each volume formulation for the product being tested in a single replication experiment. In an actual field trial, a researcher will want to either have multiple replications in a field or multiple field trials at a given testing location to ensure that any environmental effects are mitigated. In the trial design above, if the field went through a minor flooding event and the top five rows were flooded, then the researcher would have no data on low volume formulation. Luckily, they would still have some data on the positive control. One of the goals of trial design is to mitigate these effects. Depending on the size of the plot and the accuracy with which a crop protection product can be sprayed, the researcher may choose to put control plots in between the testing rows.

3 Underestimating Field Trial Costs

The cost of contracting a grower to execute a field trial includes land rental, labor, planting, monitoring, measurements, data upload, reporting, and harvesting. There are several useful calculators for this:

University of Maryland Calculator
University of Illinois Crop Budget

A contract grower who is engaged to conduct a field trial will expect a guaranteed payment premium over what they would otherwise expect if they grew a commercial crop. For this reason, it is much more costly to run a single acre tomato field trial in California than it is to conduct the trial of one acre of corn in Illinois. A basic guidance for budget building purposes is to look at average yields in the area and budget a 10-25% premium payment for a field trial.

Cost of Development

—

From 1995 to 2019 the cost of conducting the needed scale-up production, field trials, and product registrations increased from $80M to $175M (including the cost of registration in both the EU and U.S.) [1].

Trend 1

Most of this growth in cost came from expansion of the requirements for field trial performance and proof of environmental safety. Since 2019 there has been continued expansion in the number and amount of testing and trialing needed for product registration in the EU and U.S. This is inclusive of the environmental and animal safety requirements, indicating a larger growth of cost for registration. Taking a modest 15% assumption in cost growth since 2019, this would only be about half the growth rate seen between 2010-2019 and would translate to **$6M in additional cost.**

Trend 2

Field trial results must be conducted not only in a statistically significant manner. Results also much be above reproach, and engaging with 3rd party farmers, non-profits, and large institutional ag organizations will help ensure that. This may require conducting extra field trials to prove efficacy and to gain the support of the broader retail and ag industry so that when the product is ready for market there are customers willing to adopt the product. Conducting partnership field trials can add an additional season or two to the development timeline before entering the market. Taking 7 years as the standard testing period, and using the 2014-2019 numbers, this would mean ~$8M/year in field testing costs. To account for the cost of an additional year's worth of testing with partners an **additional $8M in cost is added here.**

$$\$\mathbf{216}\boldsymbol{m} = [\underbrace{\$175m}_{\text{(Reg Trials)}} + \underbrace{\$6m}_{} + \underbrace{\$8m}_{\text{(Field Test)}}] * (1 + 2.73\%)^{7}$$

Over the next few years, the use of computational and statistical models will improve our field trial design, offsetting some of the increased cost, but if the trend of registration trial and environmental chemistry requirements continues as it has historically, costs are expected to increase in this category.

This leads to the estimated cost of Phase III – Phase V: Development to range from $194M-238M.

Summary for Crop Protection Small Molecules

Small molecule crop protection products can take a long time to bring to market and cost hundreds of millions of dollars to develop. However, they represent some of the most impactful agricultural products on the market today because of their specificity, scalable production, and ease of use.

Cost Summary	Estimated Time to Market
Research **$118 - $143m** **Development** **$194 - $238m**	**Research** **3 - 5 years** **Development** **7 - 8 years**
Total Cost **$312 - $381m**	**Total Time** **12+ years**

	Research $119 - $143 m ~ 3-5 years			**Development** $194 - $238m ~ 7-8 years		
	Phase 0	**Phase I**	**Phase II**	**Phase III**	**Phase IV**	**Phase V**
	Define the Problem	Pre-field Discovery	Early Product Development	Advanced Product Development	Pre-Launch Preparation	Launch & Market Expansion
Product	**Active Ingredient**			**Formulation**		
EFFICIACY						
Hit Finding & Screening Lab, GH	– Define crop & target - Map market size - Finish customer interviews	– Refine the target - In silico, in vitro, and in vivo testing - Identify lead compound(s)				– Use field results to screen for novel targets or compounds
Lead Optimization Lab, GH			– ADMET/Eco Tox screens run – Screen for efficiacy in GH			
Field Trials Field				– 10's of acres (in aggregate) – Prove efficiacy against null hypothesis and positive control groups – Implement partner engagement strategy	– 100' of acres (in aggregate) – Test in multiple soil types, weather, and agronomic practices – Test with industry partners – Tank mix analysis	– 1000' of acres (in aggregate) – Broaden field trials to new regions for potential market expansion
Scale-Up Production Chemistry Lab, Pilot, Manufacturing Site		– Check chemical synthesis scalability	– Conduct COGS assessment of initial formulation	– Establish scale-up chemistry process for active and inert ingredients – Access or build pilot production facility	– Build or contract manufacturing for active and inert ingredients and any precursors	– Continue to seek cheaper chemical synthesis and process options to drive down COGS
Formulation Lab			– Develop initial formulation – Finalize formulation "type" for product	– Refine the formulation for efficiacy and safety – "Lock-in" formulation to be submitted to regulators		– Continue to pursue formulations that further improve product performance
Safety, FTO, and IP Field, Specialized animal and environmental testing facilities		– Conduct IP review – Establish FTO and develop regulatory and IP strategy	– Begin to execute regulatory and IP strategy – Early regulatory testing for toxicology & environmental testing including residue and metabolism analysis	– Generate data for dossier(s) to regulators	– Provide supporting data for dossier(s) – Finalize go-to-market plan – Develop stewardship plan	– Continue to provide data regulating agencies

Crop Protection Small Molecule Opportunities & Pitfalls

Hit Finding & Screening	Lead Optimization & Formulation	Scale-up Production Chemistry	Field Testing & Registration Trials
In Silico ADMET screening **Overemphasizing virtual screening** **Production costs of any chemistry can become the limiting factor when manufacturing at scale** **Failure to conduct early IP review and develop an IP strategy**	**Monitor regulatory trends** on microplastics and other formulary ingredients for evergreen compliance **The mixture challenge** **Volumes of application.** This is paramount as spray systems move toward precision application and require low, very low, and ultra-low volume formulation	**Diversify the production supply** **Sourcing more sustainable starting materials** **Reaction byproducts and heat must be managed** **Trust but verify** with builders	**Engage famers and support organizations** **Missing Positive Controls** **Underestimating Field Trial Costs**

Product Pipeline Map (Detailed)

Research
(~$119-143M) Phase 0, Phase I, Phase II

Hit Finding and Screening
Phase 0, I

Facilities
Labs
Growth Chamber
Greenhouse

Cost Trend
Decreasing due to the use of artificial intelligence to better simulate molecular binding of small molecules to targets of interest, improved screening techniques

Target identification
- Genome-wide association studies (GWAS) or sequence analysis for target
- X-Ray Crystallography or CryoEM
- In vitro & vivo assay development

Compound Screening
- Structure-based or ligand-based molecule design and dynamics prediction for affinity and library screening against target
- Active ingredient synthesis
- In vitro and vivo assay testing

Lead Optimization & Formulation
Phase I, II

Facilities
Lab
Growth Chamber
Greenhouse
Small Plot & Field Trials
Specialized animal & environmental testing facilities

Cost Trend
Increasing due to growing toxicology and environmental testing requirements; however, these are partially offset by novel in lab testing methodologies

Molecular optimization for efficacy
- Pharmacokinetic and physical characterization of leading hits and MoA
- Design for stability, efficacy, and selectivity

Formulation development
- Formulation screening trials of A.I. with inactive ingredients (wetting agents, disintegrating, diluents, fillers, binders, etc.)
- Optimization of particle size, pH polymorphism, solubility, and viscosity

Early Regulatory: Toxicology & Environ. testing
- Mammalian acute and beginning sub-chronic
- Environmental and residue analysis
- Metabolism analysis and safety assessment

Development
(~$194-238M) Phase III, Phase IV, Phase V

Scale-up Production Chemistry
Phase III, IV, and V

Facilities
Lab
Manufacturing pilot facility

Cost Trend
Increasing due the cost of raw materials needed for facility buildouts or contracts for CMOs.

Formulation and A.I. Finalization
- Synthesis route optimization
- Formulation shelf-life stability and dispersion optimization
- Tank-mix analysis

Scale-up Manufacturing
- Pilot facility scale-up for synthesis of key active and key inactive ingredients
- Build or engage CMO for scale-up production to test the pilot

Field & Registration Trials
Phase III, IV, and V

Facilities
Lab
Field Trials
Specialized animal & environmental testing facilities

Cost Trend
Increasing due to the growing requirements for registration, efficacy, and environmental trials.

Wide-scale field trials
- Testing in combination with multiple soil types, weather conditions, and agronomic practices (e.g. irrigated vs. non-irrigated)
- Side-by-side against the market standard

Registration Tests & Field Trials
- Testing for plant, mammalian, and bird metabolism and toxicology
- Testing residue lifecycle in soil/water
- All other testing needed for regulatory approval

Crop Protection
Biomolecules

New Chapter

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Introduction

Definition of Biomolecules

"Biomolecules" can be defined as carbon-based compounds found in living organisms for use in biological processes. They are composed primarily of hydrogen, oxygen, nitrogen, and carbon, and may also contain other elements such as phosphorus and sulfur. Biomolecules include the **Oligos & Peptides** and **Proteins** categories defined in the Crop Protection Product Nomenclature earlier in this Playbook, but more specifically comprise:

- Small natural molecules, such as metabolites, hormones (e.g., auxin), and vitamins
- Peptides (2 to 50 amino acids)
- Proteins (50+ amino acids), including antibodies and enzymes
- Oligonucleotide molecules (RNA, DNA)
- Carbohydrates (e.g., sugars)
- Lipids (e.g. oils, fatty acids, etc.)

Crop protection molecules type segmentation by size (in Dalton)

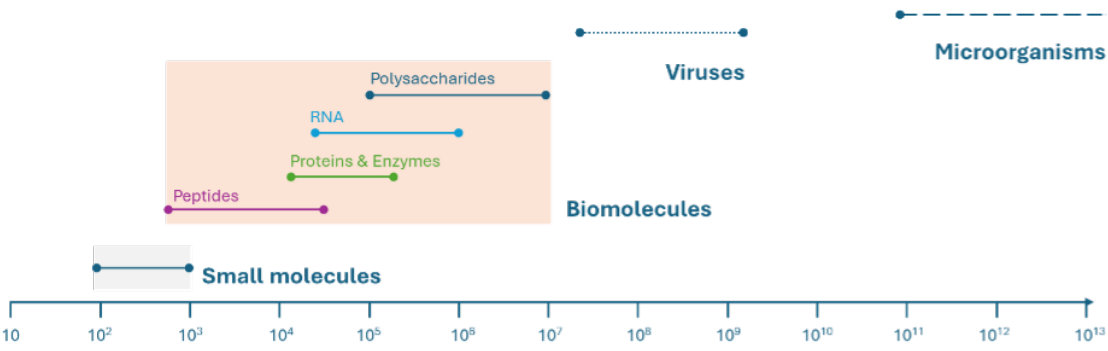

Figure 1: Crop Protection Molecules by Size

Biomolecules degrade through many different processes. Organisms break down these compounds for metabolism, growth, energy, the recycling of building blocks, detoxification, defense, and/or regulation of biological activity.

Biomolecules are used in other industries such as pharmaceuticals and cosmetics and are of increasing interest in agriculture as crop-protection products. Biomolecules represent an expansive universe of potential compounds with new modes of action (MoAs).

For simplicity, this chapter will focus on two main biomolecule categories: **amino-acid based** (peptides and proteins) and **nucleotide-based** (DNA and RNA) molecules.

It is worth mentioning metabolites, a category of biomolecule that is produced by living microorganisms (also known as microbes). The R&D steps involved in metabolite-producing microbes (amongst others) will be covered in a different chapter of the Playbook.

Similarly, pheromones are also excluded from this chapter of the Playbook. Pheromones, kairomones, and other attractants often called “semiochemicals”, are small-molecule products used to communicate or elicit a behavioral or physiological response. The specificities of pheromones in terms of MoA, screening, discovery, and manufacturing, render this class of crop protection product beyond the scope of this chapter.

Peptides, Proteins, Enzymes, Oligonucleotides

Amino Acid-Based Biomolecules

There are 22 proteinogenic amino acids, which can be combined to form longer molecules. Chains form when the carbon atom of one amino acid binds a nitrogen atom from another amino acid, known as a peptide bond. While there are hundreds of amino acids that exist in nature there are 20 ‘standard’ amino acids with selenocysteine and pyrrolysine making up the 21st and 22nd on the proteinogenic list.

The term “Peptide” usually refers to short chains of ~2 to 50 amino acids, whereas “protein” is used for longer chains above 50 amino acids. The longest known protein today is made of 34,000 amino acids (PKZilla).[22]

Based on their amino-acid composition and length, peptides and proteins come in many sizes and shapes. Some chains are linear, others are cyclic, and their myriad three-dimensional structures offer a huge variety of properties.

Peptides and proteins can serve many different functions including but not limited to antimicrobial properties, signaling, or catalysis (enzymes), which facilitate chemical reactions.

Beyond the 22 proteinogenic amino acids, scientists have developed synthetic amino acids to improve the chemical properties of natural peptides (e.g., stability, enzymatic kinetics).

However, using synthetic amino acids can pose other challenges in terms of large-scale manufacturing and extending the regulatory pathway. Regulatory agencies may not recognize these products as identical to natural products.

[22] https://www.fiercebiotech.com/research/largest-known-protein-has-been-discovered-algae-and-its-machine-makes-poison

Nucleotide-Based Biomolecules

Nucleotides are the basic building blocks of nucleic acids (RNA and DNA). They are made of a sugar molecule attached to a phosphate group and a nitrogen-containing base. RNA molecules are composed of four nucleotides: adenine (A), uracil (U), cytosine (C), and guanine (G); DNA is composed of adenine (A), cytosine (C), thymine (T), and guanine (G).

Various types of RNA molecules play unique functions in living cells: messenger RNA (mRNA), microRNA (miRNA), transfer RNA (tRNA), etc. The main RNA function leveraged for crop-protection applications is RNA interference (RNAi). With RNAi, an RNA molecule (usually either double-stranded [dsRNA] or small interfering RNA [siRNA]) binds to its target mRNA to inhibit its function.

The most used RNA-based biopesticide type is dsRNA, because it combines relatively high stability (compared with single-stranded RNA [ssRNA]) with the specificity of targeted RNAi-mediated gene-silencing. This works via an enzyme called Dicer, which cleaves the dsRNA into siRNAs. These siRNAs then assemble into an RNA-induced silencing complex (RISC), which affects specific mRNA degradation in the target pest, pathogen, or weed.

An Untapped Universe of New Modes of Action

There are two approaches to biomolecule discovery. Examples below will highlight how these can unlock a new universe of potential products.

Target-based

- Examples: RNAi, antibodies, effectors of protein-protein interaction (PPI)
- Start with a target (protein, RNA, other)
- Then design a biomolecule de novo, using building blocks (amino acids or nucleotides) to specifically interact with the target. While not required, this biomolecule is often similar in structure and identical in function to a naturally occurring molecule

Nature-derived and optimized

- Examples: signaling peptides, antimicrobial peptides (AMPs), neurotoxins
- These molecules are either naturally occurring, partially modified, or synthetic analogs with improved properties of interest (e.g., potency, stability)
- Modifications may be to the nucleotides or amino acids themselves, or to elements of their structure or backbone
- It should be noted that modifications to a nature-derived product can change its regulatory classification

Similarities and Differences with Small Molecules

Similarities	Differences
• Target-based and rationally designed (biomolecules interfering with expression or activity of an essential target protein) • Potentially applicable to all indications: insecticide, fungicide, herbicide • Highly specific and optimizable molecules • Varying levels of light, pH, and temperature stability • Better compatibility/mixability with other crop protection products than living comparables (e.g., microbes) • Multiple MoAs	• Different manufacturing systems: bioprocesses (in most cases) for biomolecules vs. chemical syntheses for small molecules • Size and resultant bioavailability, penetrability, translocation, and systemicity: many biomolecules are bigger than small molecules and may have more difficulties penetrating insect, fungal, or plant cells and organs • Biological stability/degradability. Biomolecules may degrade quickly in the environment with a desirable safety profile. The downside is that they may degrade too fast to have the desired effect or industry-standard residual efficacy • Faster regulatory pathway for “natural products” in some geographies (e.g., North America and South America) • The chances of off-target effects are reduced

By the numbers (disclaimer): Because there are far fewer public data points around the timing and cost of development for many of these products, the financials projected in this section have been estimated by the expert authors and reviewers of the Playbook. As with all numbers in the Playbook, these costs could be higher or lower. However, the included figures are meant to serve as a basis point or benchmark when thinking about the time and cost associated with developing crop-protection biomolecules.

Stage Gate Plan Summary

Figure 2: Stage Gate Plan for CP Biomolecules

Abbrevations:
TGAI = Technical Grade Active Ingredient; DSP = Downstream Process;
IP = Intellectual Property; FTO = Freedom to Operate;
CRO = Contact Research Organization; CDMO = Contract Development & Manufacturing Organization; CMO = Contract Manufacturing Organization

Bio-molecules	**Research** $5-13M 2 to 5 Years			**Development** $25-49M 5 to 9 Years		
	Phase 0	**Phase I**	**Phase II**	**Phase III**	**Phase IV**	**Phase V**
	Define the Problem	Pre-field Discovery	Early Product Development	Advanced Product Development	Pre-Launch Preparation	Launch & Market Expansion
Product	**Active Ingredient (Synthesis)**		**TGAI (Technical Grade Active Ingredient)**	**Formulated Product (based on TGAI)**		
Hit Finding & Screening Lab, GH	- Define Crop & target - Map market size - Finish Customer interviews - First TEA (Technco-Economic Analysis)	- Target Selection - Design-to-Hit: binding / in-vitro - Hit-to-Lead: in vivo (Glasshouse)				- Use field results to screen for novel targets or compounds
Lead Optimization Lab, GH		– Analog / variant design to overcome production / stability weaknesses	– Selectivity / spectrum – Use rate & Timing of Application – Preliminary Formulation tests			
Field Trials Field			– Semi-field trial – Greenhouse to Field translation	- ~10's of acres (in aggregate) - Prove efficacy against null hypothesis and positive control groups - Implement partner engagement strategy	- ~100's of acres (in aggregate) - Test in with multiple soil types, weather, and agronomic practices - Test with industry partners - Tank Mix analysis	- ~1000's of acres (in aggregate) - Broaden field trials to new regions for potential market expansion
Production Lab, Pilot, Manufacturing Site		– Lead to TGAI: Feasibility: Flask & 2L fermentation, inc. DSP	– Scale-up: 2L to 1m³ (x1,000) – Analytical: bi-products	– Process Development & Pilot Scale: 100L to multiple cubic meters (ex. 15m³, 75 m³) – Process Freeze – 5 Batch study – CDMO screening	- First production batches including formulation and packaging. - Technology transfer from CDMO to CMO to secure access to production capacity - Continued process optimization	- Contract with CMO signed and supply chain established for distribution
Analytical Chemistry & Formulation Lab		– Biological stability profile – Phys-Chem Properties	– Develop Initial Formulation	- Refine the formulation for efficacy and safety - "Lock-in" formulation to be submitted to regulators (Formulation Freeze)	- Formulation industrialization in parallel to TGAI. - Initiate development of additional formulations for future products based on same TGAI	- Continue to pursue formulation to could further improve product performance
Safety, FTO, and IP Field, Specialized animal and environmental testing facilities		– IP Review	– FTO and IP Strategy – Early Tox Studies (genotoxicity, etc.) – Preliminary Data Gap Analysis to prepare Regulatory strategy	- 5 batch Study - Regulatory studies preparation (CROs identified and contracted, ...)	- Finalize Reg. studies and submit dossier - Finalize the go-to-market plan - Develop product stewardship plan	- Data Review by Regulatory agency (18 to 46 months) - Approval in first markets - Continue to provide data to regulating agencies
Approximate Timing	2 to 4 years			5-8 years		
	1-6 months	~6-18 months	~18-36 months	24-36 months	24-48 months	12-24 months
Approximate Cost	$5-13M			$20-42M		
	$50-300k	$1-4m	$4-9m	$7-10m	$8-20m	$5-12m

Phase 0 – Phase II:
Research

Phase 0:
Product Concept

Timing

1–6 months

Summary of Main Goals during Phase 0

During Phase 0, the main objective is to develop a compelling business case for the launch of a new product. This business case should include three main elements:

1. An evaluation of the market potential, technology suitability, competitive landscape, and regulatory environment surrounding the target product profile
2. Translating economic targets in $/acre into R&D targets and not the reverse. Working backwards from:
   a. Target price for the farmer/customer ($/acre)
   b. Target product cost ($/acre) to retail/distributor
   c. Target use rate (g/acre) and target manufacturing cost in $/g
3. The result of this analysis should drive the R&D process in the right direction to reach unit economic goals

Opportunities & Common Pitfalls

Opportunities

- Access to the farmers/customers to conduct interviews on product need, build the financial models to understand the 'business case' and market potential for the product
- Leveraging Artificial Intelligence to support techno-economic analyses (TEAs) and market studies
- Research across the life science industries (even outside of Ag) into current best-in-class products on the market and recently published papers or patents in to ensure basic freedom to operate

Common Pitfalls

- No or wrong TEAs
- Looking at the "wrong" target: for example, focusing only on technical targets (e.g. production yield in g/L) without looking at the broader and more important economic targets ($/acre)
- Overestimating target market and potential revenues including the Target Addressable Market (TAM), Serviceable Available Market (SAM), Serviceable Obtainable Market (SOM)

Cost Range & Trend

$0.1–0.3 million

Remaining cost neutral or decreasing slightly. The cost of conducting interviews mostly remains the same, however, the growth of social platforms makes farmers and customers more accessible.

The product concept phase for the development of biomolecule-based biopesticides follows similar principles to that of chemical pesticides. Market size and revenue potential should be estimated, biological targets need to be defined regarding application (pests, diseases, weeds), crop types (high value vs. row crops), and geographies (US, Latin America, Europe, Asia). Equally important, unit economics and corresponding technical targets should be estimated and set early on to drive R&D efforts towards clear quantitative goals. Any comparable products as examples will convey important information about the viability of producing a product at scale.

As an emerging and growing product class, biomolecule crop protection products are held to the same standard as small-molecule crop-protection products, including efficacy, stability (both for shelf-life and stability in the field) and application using established agricultural equipment and practices. These products should be combinatorial with existing products, small molecules or otherwise. Finally, pricing should be competitive and comparable to chemicals.

Technology suitability is a critical consideration for biomolecule products. Generally, with a well-understood MoA and physicochemical properties, one should assess if the molecular target is "reachable" by the biomolecule. Take molecular size and permeability as an example. Whereas chemicals are frequently small enough to penetrate the targeted cells, biomolecules may not be able to do so. Verifying the suitability of the technology for the molecular target of interest represents an important validation to optimize the probability of success in controlling the target pest.

During Phase 0, it is recommended to not only look at the market potential and technical viability, but to also analyze and understand the required unit economics to conduct a first "reality check" of the feasibility of developing a new product and set the right technical and economic targets early on.

A good approach to market sizing is to combine both "top-down" and "bottom-up" methods to pressure test assumptions and come to a realistic estimate. For example, in the case of a novel bioinsecticide product for corn in the US, a top-down approach would first look at market data on the total insecticide market globally, then geographically on the first targeted market (the US) and, if data are accessible, on the target crop (corn). A bottom-up approach would consider the total cultivated area in the US multiplied by the average spend per grower on insecticide products each season ($/acre). Another important piece of market information is the number and timing of applications per growing season. Crop-protection products are often applied several times per season, with applications varying from one or two to over ten in a season depending on the crop. It is also possible to access data on existing products (chemicals and biologicals) with estimated revenues and cost or selling price ($/acre).

For example, regarding selling price specifically, a simple approach worth exploring is based on a value-sharing assumption. The basic idea is that the new product should give farmers a consistent yield increase (e.g., 5% yield increase in kg/acre), which then could be translated into value increase ($/acre on average). The next step is to take an assumption on the value repartition across the value chain between the farmer (e.g., 50%), distributors (e.g., 20–30%), and manufacturer (e.g., 20–30%). This approach can give a first estimate of the potential value captured by the new product and the corresponding selling price target (in $/acre). Pairing this type of analysis with information on competing products, particularly the current selling price per acre of the main benchmark product, will help build more robust price targets.

These first economic analyses should provide the right guidelines to the whole R&D organization towards the required price range to be able to compete in the targeted market.

The second important step in Phase 0 is to perform a reality check to see if and how the target price could be achieved, and if it is possible to achieve healthy margins. We recommend revisiting/refining this calculation at the end of each subsequent phase. Key parameters to estimate

and measure include field efficacy vs. dose rate (grams of active ingredient [AI]/acre) and estimated manufacturing costs ($/g AI). In the case of biomanufacturing, manufacturing cost is often directly linked to process yield (e.g., grams of AI per liter of fermentation). Both parameters are influenced by a variety of factors – as shown in Fig. 1.

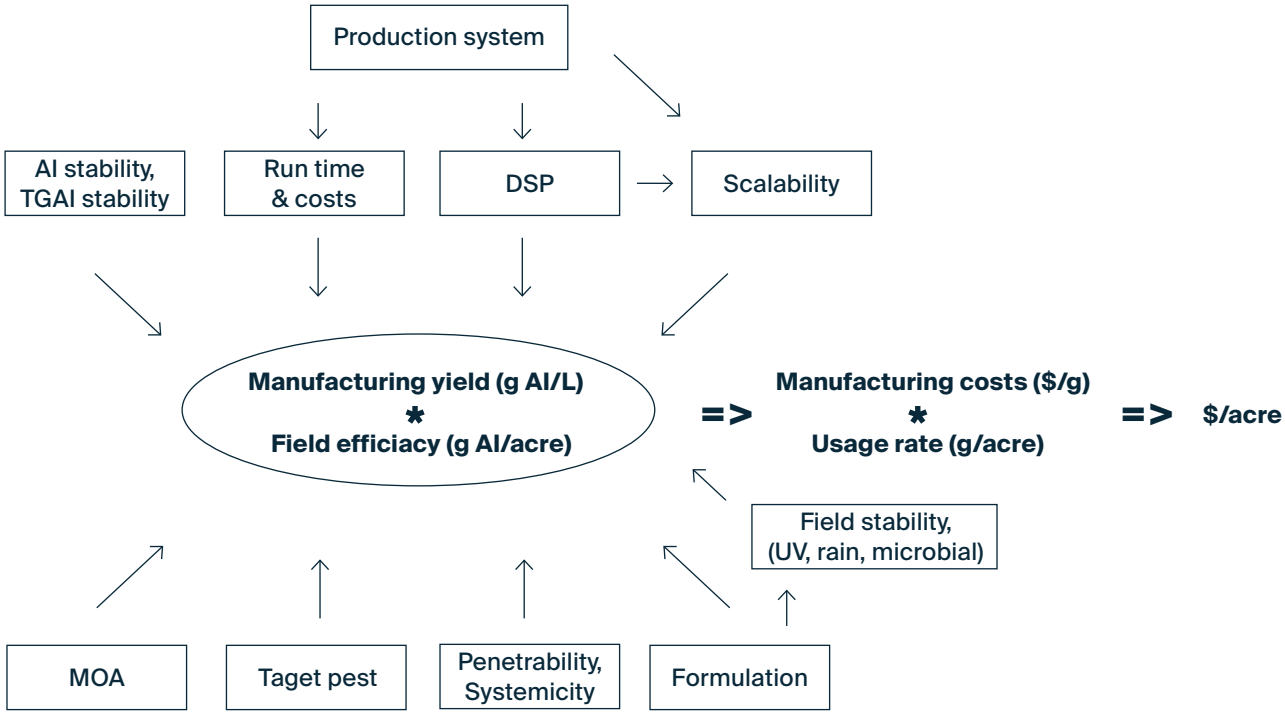

Figure 3: Pricing of Biomolecules and influencing factors

Let's take an example of a novel crop-protection biomolecule produced in a yeast-based system and potentially offered as dry formulation (e.g., water-dispersible granules, WDG) with the following assumptions:

- Target price of a novel biopesticide (price to distributor, based on existing competitors): $40/acre
- Field efficacy/usage rate (based on preliminary results): 20g/acre
- Fermentation titer (realistic for optimized yeast-based manufacturing): 15g/L
- Manufacturing costs: $10/L = $10/15g of AI = $0.67/g AI
- $0.67/g × 20g/acre ≈ $15/acre

In this example, cost of goods sold (COGS) of $15/acre seems possible and a target price of $40/acre to distribution partners financially viable.

Depending on the MoA, usage rates of less than 10g/acre are possible (e.g., for some RNAi products). Usage rates of more than 200g/acre on the other hand require very low manufacturing costs to be economically viable. Higher usage rates can also lead to greater regulatory review

timelines. The results from these initial manufacturing analyses should inform the organization about the future fermentation volumes that will be required for the targeted product. Figure 4 below provides a high-level view of the different steps required to reach the commercial manufacturing process and the associated costs of goods. Please note, this graph is generalized, and the range may vary with different technologies (e.g. catalytic products like RNAi or enzymes may require lower volumes due to smaller application rates); it helps to understand the key steps leading to manufacturing at scale.

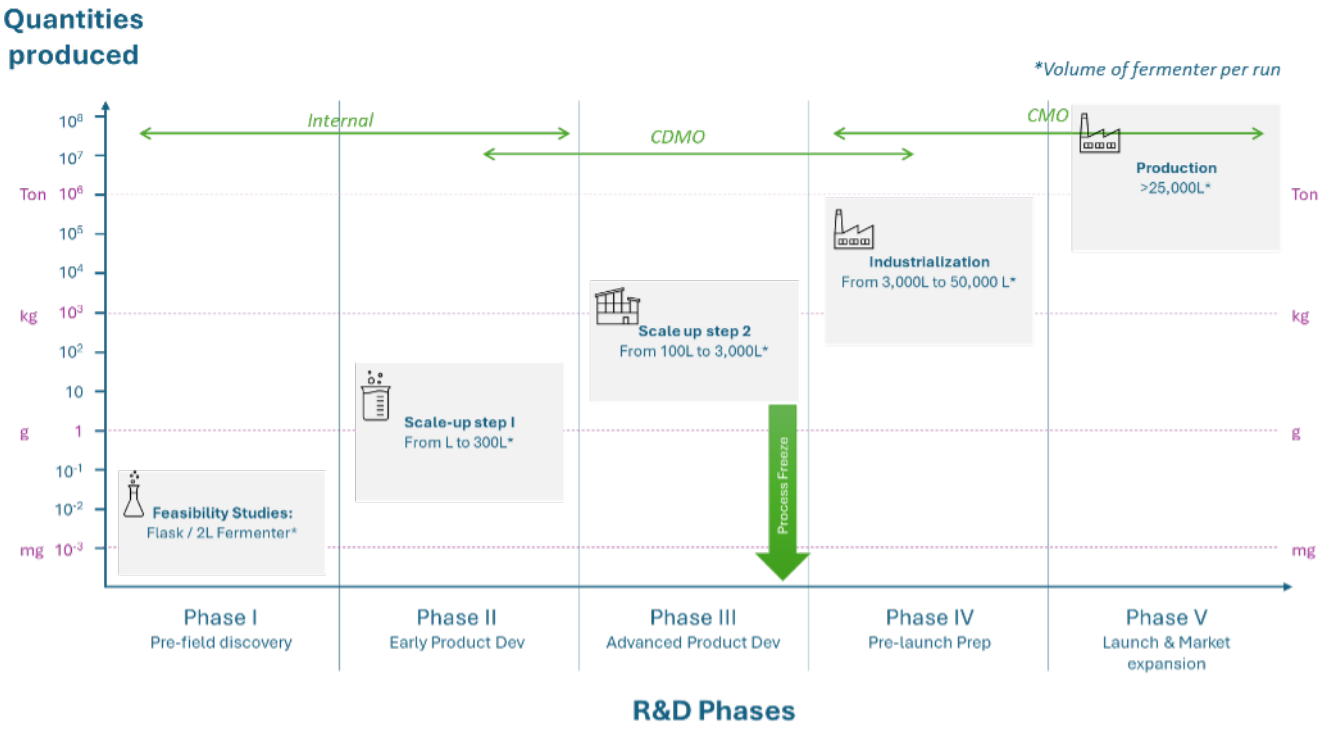

Figure 4: Scale-up Production Phases

While the organization will probably have little technical data at this stage, simulating what it would take from a cost of manufacturing perspective to achieve the desired economic target is a must-do. For example, based on the case described above and with a target price set at 40$/acre, a distributor margin of 30% and a gross margin of 50%, the R&D team will need a new AI providing efficacy in the 10–30g/acre range and with a biomanufacturing process performance of 10–25g/L.

1. Economic Targets & Assumptions

Farmers price	$/acre	40
Distributor margin	$/acre	12
Product price to distributor	$/acre	28
Target growth margin	%	50%
Target product cost	$/acre	14

2. Corresponding Technical Targets

Production Cost $/L		10	10	10	10	10
Process Yield g/L		5	10	15	20	25
Product Cost $/g		2.00	1.00	0.67	0.50	0.40
Use Rate (g/acre)	10	20.0	10.0	6.7	5.0	4.0
	20	40.0	20.0	13.3	10.0	8.0
	30	60.0	30.0	20.0	15.0	12.0
	40	80.0	40.0	26.7	20.0	16.0

Figure 5: Example unit economics for a biomolecule

Biomolecules can offer benefits to growers, consumers, and the environment that crop-protection small molecules often cannot provide. Take the critical elements of environmental safety and user safety, for example. Biomolecules degrade into natural building blocks, such as amino acids and nucleotides, meaning their environmental fate is rarely problematic. Non-toxicity for humans and non-target organisms is not an intrinsic feature of all biomolecules, but they often express favorable safety profiles based on specificity and MoA.

However, this specificity that makes the biomolecule safe, can also limit its market potential. For example, a bioinsecticide that is safe for off-target organisms like honeybees and other pollinators may not be active against all target pests. A bioherbicide may only be active against a narrow range of closely related weeds. These potential limitations should be considered when defining the product concept and estimating its potential return on investment (ROI).

Another benefit of biomolecules is the potential to act through a novel MoA and overcome the increasing resistance challenges that crop-protection small molecules are facing in the field. This has been one of the biggest challenges for the crop-protection small molecule industry over the past decade. A novel MoA is desirable for any new pesticide and should be a “must” for newly developed crop-protection biomolecules. While there can be multiple sites of action (SoA) to a MoA, only a new MoA resets the resistance clock and provides a basis for a long product life cycle.

Phase I:
Pre-field Discovery

Timing

6–18 months

Summary

The main goal during Phase I is to design or discover peptide, protein, or oligo-based molecules and rapidly evaluate their efficacy, delivery feasibility, cost of production, and biological stability. This early stage serves to eliminate unviable options and establish a shortlist of candidates worth advancing into further development. Candidate molecules take on various titles during this period:

- "Hit" candidates have shown good level of activity in vitro after a first selection process
- "Lead" candidates have shown good level of activity *in planta* against the targeted pest or pathogen
- "Technical Grade Active Ingredient" (TGAI) lead candidates have been successfully produced using a lab-scale version of the future manufacturing process (bioproduction or synthesis), have shown good stability and penetrability properties (if the MoA requires it), and have shown a good level of activity *in planta*

During Phase I, candidates move from design to TGAI and provisional IP should be secured for key innovation

Opportunities & Common Pitfalls

Opportunities

- Leveraging development in artificial intelligence to speed up and improve the accuracy of biomolecule design
- Access to the relevant "omics" data for target selection and design (e.g., access to annotated genome, protein target already identified and modeled)
- Access to phenotyping capabilities (binding assays, in-vitro or in-planta assays) instead of taking more time to develop your own bioassay
- Access to the appropriate biomanufacturing technology for the relevant candidate or choosing to pursue custom development

Common Pitfalls

- Improper Selection: Promoting the "wrong" candidates to the next stages, requiring projects to start over
- Overly selective: Promoting only one candidate to Phase II without having at least one or two backup options
- Confirmation bias: Spending too much time trying to optimize a candidate that is not going to make the cut (e.g., very active but impossible to produce)
- Analysis paralysis: Spending too much time trying to optimize everything and not knowing to stop when it is "good enough"

Cost Range & Trend

$1–4million

Cost remaining neutral as in silico and in vivo testing throughput capabilities and number of compounds continue to grow, but still require biological assays to verify

Cost will also vary based on the complexity of the targeted phenotype. For example, measuring nutrient absorption can be more complex than measuring insect death, which would require more time to screen the same number of biomolecules

Introduction:
Phase I Biomolecules

This section will explore two approaches for discovery of crop-protection biomolecule products: **target-based design** and **nature-derived products**. In each approach Phase I discovery proceeds through three steps:

Step 1.1: Hit Identification (Design-to-Hit or Sourcing-to-Hit)

- **1.1.1 Target Selection and Biological Rationale**
- **1.1.2 Molecular Modeling and Structure Prediction**
- **1.1.3 In-Silico Starting Point: Docking Simulations and Interaction Scoring**
- **1.1.4 Biomolecule Validation**
- **1.1.5 In-Vitro Validation**

Step 1.2: Lead Optimization

Step 1.3: Lead-to-Technical Grade Active Ingredient

- **1.3.1 Production Route Feasibility Studies**
- **1.3.2 Efficacy Validation of the TGAI**
- **1.3.3 Stability and Penetration Studies**
- **1.3.4 Lead Sequence Optimization and MoA Validation (Optional)**

Target-Based Design (Design-to-Hit)

In this approach, molecules are designed de novo. The discovery process begins with a specific biological target—such as a gene, receptor, or enzyme—and researcher(s) design biomolecules (dsRNA, peptide, protein, etc.) to interact with or disrupt that target. This approach shares significant conceptual similarity with chemical crop protection.

Examples of target-based design technologies:

Technology	Targets	Description	Examples
RNAi	mRNA	Delivers dsRNA that is processed into small RNAs (siRNAs), which bind to specific mRNA in pests or pathogens and prevent protein production by destroying the mRNA	Sprayable dsRNA against Colorado potato beetle or fungal pathogens and weeds
Antibodies, Nanobodies	Proteins (in pathogens or pests)	Bind very specifically to a target protein (fungal enzyme or toxin, or an essential enzyme/protein in a pest), blocking its activity or marking it for destruction	Antibodies/nanobodies that neutralize fungal virulence factors or block proteins in the insect gut
Peptide–Protein Interaction	Proteins (host or pathogen)	Small peptides interfere with protein–protein interactions by binding to a protein and blocking its function or preventing it from interacting with other molecules	Synthetic peptides blocking fungal signaling or infection pathways
miPEP (microRNA-Encoded Peptide)	miRNA (regulatory RNA)	Small peptides encoded by the same gene as miRNA that **enhance the expression** of that miRNA, amplifying its natural gene-regulating effects	miPEPs used to enhance stress tolerance or disease resistance in plants
Enzymes (Proteins)	Cell walls, proteins, or molecules in pests/pathogens	Act as biological scissors or glue that either **cut/degrade** or bring together specific molecules leading to death or weakness of the pest/pathogen	Chitinases degrading fungal cell walls; proteases disrupting insect gut lining

Nature-Derived Discovery (Sourcing-to-Hit)

This approach involves identifying biologically active peptides or proteins from natural sources such as plant extracts or microbial metabolites. These biomolecules are either used in their native form or further optimized for agricultural use.

Examples:

Technology	Targets	Description	Examples
Signaling Peptides	Plant receptors and signaling pathways	Mimic natural or pathogen-associated peptides to **trigger plant immune responses**, preparing the plant to fight off real threats	Flagellin, Harpin proteins, or derived synthetic peptides
Insecticidal Peptides	Ion channels or membranes in insect pests	Bind to and disrupt **critical proteins in insect nervous systems or membranes**, causing paralysis or death	Pyrokinins, spider venom peptides, ant venom peptides
Antimicrobial Peptides (AMPs)	Bacteria or fungal membranes or proteins	**Disrupt cell membranes, degrade cell walls**, or **block essential functions** in bacteria or fungi, leading to their death	Plant-expressed AMPs targeting fungal or bacterial pathogens
Hydrolysates (Proteins or Peptides)	Plant metabolism, receptors, or defense pathways	**Stimulate plant growth or defenses** through bioactive protein/peptide fragments taken up by the plant; often act as biostimulants or primers of immunity	Protein hydrolysates promoting growth or resistance to stress

1.1 Hit Identification (Design-to-Hit or Sourcing-to-Hit)

A. Target-Based Design Approaches: Design-to-Hit

The goal of the **targeted approach** is to rationally design and iteratively refine a biomolecule (e.g., dsRNA, peptide, or protein) that binds to a specific biological target and blocks, activates, or modulates its function. Candidate biomolecules are put through efficacy validation, ensuring that the theoretical interaction translates into meaningful biological impact under realistic conditions—first in vitro, then in model systems, and ultimately in plants or pests.

There are five main steps to this approach:

- **Target selection:** Identify an RNA or protein that is critical for pest survival, plant defense, or pathogen virulence.
- **Molecular modeling:** Predict the structure of said RNA or protein using tools like AlphaFold or RNAfold.
- **In-silico docking simulations:** Leverage computational tools (e.g., AutoDock, Rosetta, machine-learning-based siRNA efficacy prediction models) to simulate interactions between designed molecules and their targets.
- **Biomolecule validation:** Conduct binding assays, gene silencing efficiency (for RNAi) assays, or enzyme inhibition assays to validate activity of your candidate biomolecules.
- **In-vitro assays:** Assess the efficacy on the targeted pathogen or pest by measuring its IC50, EC50, etc. (Please see the definitions of IC50 and EC50 in the Crop Protection Small Molecule section of the Playbook.)

Let's take a deep dive on each of these different steps:

1.1.1 Target Selection and Biological Rationale

The first step is to identify a biologically relevant target:

- In **insects**, this could be a gene essential for growth, reproduction, feeding, or survival (e.g., V-ATPase, chitin synthase, Dicer).
- In **pathogens,** this could be a virulence factor, membrane protein, or essential enzyme.
- In **plants**, this could be a regulator of stress response, development, immunity (e.g., transcription factors, miRNA), or an essential metabolic step depending on the targeted phenotype (e.g., herbicidal activity, drought resistance, nutrient absorption).

Selection of a biologically relevant target is often driven by:

- Gene expression profiles using RNA-sequencing to know if and where the target is expressed
- Genetic essentiality screens using CRISPR or RNAi libraries
- Conservation across pest populations
- Availability of 3D structural data
- Mutant collections containing relevant phenotypes

For example, in RNAi applications, the target must be expressed in accessible tissues (e.g., insect midgut epithelium), responsive to dsRNA uptake and susceptible to dsRNA-mediated silencing.

Nanobody Example:
Intelligence in Immune Response

Nanobodies (recombinant variable domains of heavy-chain-only antibodies [HcAbs]), although naturally found in camelids and sharks, can be specifically triggered by injection of a molecular target or a portion of it. The animal's immune system then generates millions of antibodies against the target (e.g., fungal or insect) and high-throughput screening can identify the most promising antibodies. However, only the nanobody is of interest as an AI; through further analysis and encoding into a vector, it can then be expressed in a host cell. Nanobodies have a molecular weight of roughly 15kDa, approximately one-tenth of the molecular weight of conventional antibodies, making them the smallest known natural antibodies. Nanobodies are structurally different from traditional antibodies, showing higher affinity and stability, microbial expression, low immunogenicity, good water solubility, and strong tissue penetration.

1.1.2 Molecular Modeling and Structure Prediction

Once a target gene or protein is selected, computational tools are used to model its structure and interactions:

- **For proteins**, homology modeling (e.g., using AlphaFold2, SWISS-MODEL) predicts the 3D conformation of the protein.
- **For RNA**, RNAfold and Mfold models secondary structures to predict accessible regions for RNAi targeting.

These models guide where and how to design interacting biomolecules—whether an **RNA sequence** (for silencing), a **peptide** (for binding), or an **antibody** (for neutralization).

1.1.3 In-Silico Starting Point: Docking Simulations and Interaction Scoring

Docking simulations test the fit between a designed molecule and the target's active or allosteric site(s):

- **Molecular docking software** (e.g., AutoDock, Rosetta, HADDOCK) simulates how the peptide or RNA segment will interact with the target's binding site of interest. Parameters like **binding energy, hydrogen bonding, solvent exposure, and electrostatic fit** are analyzed. For RNAi, tools like **siDirect, DSIR**, and **RNAxs** optimize siRNA/dsRNA sequences for accessibility, specificity, and minimal off-target effects.
- Many of these tools are still being improved for applications to agricultural species of interest (plants, microbes, insects, etc.)

These in-silico methods enable the design of high-affinity molecules before any wet-lab testing.

1.1.4 Biomolecule Validation

Selected candidates should then undergo **biomolecule testing** to confirm activity on target of interest:

For peptides/proteins:

- **Binding assays** like Enzyme-Linked Immunosorbent Assay (ELISA), Surface Plasmon Resonance (SPR), and Microscale Thermophoresis (MST) test interaction with the target.
- **Enzymatic inhibition assays** if the target is an enzyme.

For RNA:

- **In-vitro transcription** of dsRNA followed by exposure to cell lines or pest tissues.
- **Quantitative Polymerase Chain Reaction (qPCR) or western blot** to measure knockdown efficiency at the RNA/protein level.

1.1.5 In-Vitro Validation

Selected candidates also undergo in-vitro **testing** to confirm activity on the targeted phenotype (fungal control, insect control, etc), for example:

- EC50 for fungicidal activity as example
- IC50 for insecticidal activity as example

In-vitro testing can usually be completed in systems with medium to high-throughput to rapidly validate the biological activity of the selected candidates

This step filters out inactive candidates to yield a smaller set of validated candidates. The best candidates identified at this stage are qualified as “hits”.

B. Nature-Derived Molecules: Sourcing-to-Hit

The first step **(discovery/identification)** of the discovery process is different for biomolecules that are derived from natural sources. In this case, the process starts with identifying a specific peptide, protein, or oligonucleotide molecule, or a specific family of these that share the same functionality and properties. Companies can identify these natural biomolecules through different means: internal research programs, technology scouting and scientific watch, collaboration with academic partners, etc. Libraries of these biomolecules then go through in-vitro screening in the form of assays against pests, pathogens, or plants to evaluate activity (as in section 1.1.5).

The second step **(material procurement)** revolves around ways to source the material, with two main approaches: 1) extracting the biomolecule from a living source (e.g., hydrolyzation) or 2) synthesizing the core amino-acid or nucleotide sequence of interest. Biomolecule synthesis has made large technological strides in the past decade, enabling cheaper procurement and the ability to test a larger number of potential candidates, however, it is important to understand whether costs of production will scale to commercial levels.

Although the first steps differ from the de-novo design approach, the outcomes at the end of 1.1 are the same: the company should have qualified hit candidates validated with in-vitro efficacy data. The next step is to advance the selected hits into in-vivo assays to identify “lead candidates”.

1.2 Lead Optimization

As with lead optimization of small molecules, this stage focuses on taking hits from stage 1.1 (design-to-hit or sourcing-to-hit) and optimizing their physical and chemical properties for absorption, binding, and speed of response. During lead optimization, the assays will move from in vitro to in vivo and whole-plant studies.

The goal and outcome of lead optimization is to move the hits through in vivo assays in planta to qualify them as leads, which would then be optimized in formulations and tested in the field.

Efficacy assays at this stage would typically be conducted on whole plants in a greenhouse or growth chamber and against the real target (weed, insect, or fungal pathogen). During this stage, the team can also start experimenting with different use rates, number of sprays, and timing of application to better assess the effectiveness of each hit to qualify the best ones for the next stage.

1.3 Lead-to-Technical Grade Active Ingredient

This stage can be broken down into four main steps:

- Production route feasibility studies
- Efficacy validation of TGAI
- Stability and penetration
- Optional: Sequence optimization and MoA confirmation

1.3.1 Production Route Feasibility Studies

For peptide, protein, and oligonucleotide crop-protection products, selecting the appropriate **production route** is a critical early decision. Unlike crop-protection small molecules, which are typically made via chemical synthesis, most biomolecules are too large, complex, or unstable to be synthesized economically at scale using traditional chemistry. This is where **biomanufacturing processes** become essential. As the saying goes, for biomolecules and biologicals alike “process is the product”. Once a hit has become a lead with a robust level of efficacy, the researcher/company should rapidly begin work on the technical grade version of the product before moving too far through the R&D phases.

It is important to switch early on from research-stage production of pure AIs (e.g., *Escherichia coli*-based or chemical synthesis) to a commercially viable manufacturing system (e.g., yeast or *Bacillus* based) that produces TGAI. Determination of the form of the TGAI between a dry formulation or a liquid concentrate should also be made as soon as possible. Each formulation has pros and cons; the choice is often determined by the biological stability of the AI, preferred customer experience, and early-stage results in small-scale field conditions.

Chemical Synthesis vs. Biomanufacturing

- Chemical synthesis can be used for small molecules such as small peptides (below 20 amino acids). These small peptides can be synthesized chemically at large scale with an estimated cost of goods around $100/g. This approach could be economically viable if field doses are very low (0.01–1g/ha).
- While there have been improvements in the de-novo synthesis of some biomolecule classes, biomanufacturing is usually needed for peptides/proteins or oligonucleotide molecules requiring doses higher than ~1g/ha. Chemical synthesis being either too complex (e.g., long molecules, need for specific refolding) or too expensive.

What is Biomanufacturing?

Biomanufacturing refers to the use of living cells (or biological cell-free in-vitro systems) to produce complex biological molecules. These cells act as microscopic factories, programmed to synthesize the desired peptide, protein, or RNA sequence. The final product is then extracted and purified for agricultural use.

Biomanufacturing is more than just fermentation; it encompasses:

- **DNA vector design**. Introducing the gene coding for the active biomolecule into the host.
- **Expression system engineering**. Ensuring the host can efficiently transcribe and translate the gene.
- **Fermentation** or **“upstream process”** (USP). Growing the engineered host in bioreactors to accumulate product in a batch, fed-batch, or continuous (submerged liquid) fermentation process.
- **Purification** or **“downstream process”** (DSP). Isolating the active biomolecule from other cell components through various techniques including but not limited to:
  - Separation. Such as precipitation and re-solubilization).
  - Concentration. Various filtration technologies including tangential flow-filtration, micro- and ultra-filtration.
  - Drying. Spray drying, fluidized bed drying, etc.

  An efficient and cost-efficient DSP is often the most critical step to achieve a cost-of-good position compatible with the crop-protection market.
- **Analytical sciences.** To confirm the presence and quantify the targeted lead molecule in the final TGAI and identify potential by-products. **The importance of accurate and robust analytics should not be underestimated. Failing to establish these in Phase I, will lead to misinterpretation, delays, and extra costs during later stages of development.**

The choice of the best bioproduction system depends on several factors:

- Size and complexity of the molecule
- Post-translational modification needs
- Dosage and cost targets
- Stability and formulation constraints

Main Biomanufacturing Systems for Peptides, Proteins, and RNA in AgTech

There are five main categories of biomanufacturing systems available for peptides, proteins, and RNA:

- **Yeast** expression systems
- **Bacterial** expression systems
- **Filamentous fungi**–based expression systems
- **Cell-free** systems
- **Other systems including**: molecular farming, field-grown production, solid-state fermentation

Let's take a look at each one of them from a production perspective (USP) before briefly looking at the DSP.

Yeast Expression Systems

Yeast are eukaryotic single-celled fungi and are often the system of choice for producing peptides and proteins that require **basic post-translational modifications** (like disulfide bond formation or glycosylation).

How it works

- A gene (or multiple genes) encoding the target peptide/protein is inserted into a yeast-compatible vector.
- Yeasts are transformed with this vector and grown in fermenters (1L to >10,000L scale).
- Production strains contain multiple genomic insertions of the gene-of-interest to achieve a high-level production and to ensure strain stability.
- The recombinant product is secreted into the culture medium, which simplifies purification.

Common species

- *Pichia pastoris*. Favored for high-yield secretion of recombinant proteins.
- *Saccharomyces cerevisiae*. The classic baker's yeast, well characterized but less efficient for high-yield protein production than Pichia.
- *Kluyveromyces lactis*. An alternative to P. pastoris with similar growth rates, excellent protein folding (including disulfide bond formation), and secretion properties.

Advantages

- High biomass growth rate
  Scalable and cost-effective
- Capable of simple folding and disulfide bond formation
  GRAS (Generally Recognized as Safe) status for some species

Limitations

- Less suitable for complex glycosylation or large proteins (>50 kDa), in particular, if a specific glycolysation pattern is crucial for the activity, specificity, or stability of the protein of interest.
- Proteolytic degradation may occur (protease-deficient strains may reduce this problem)

Bacterial Expression Systems

Bacteria (especially E. coli) are widely used due to their simplicity, fast growth, and low-cost media. They're ideal for **short peptides and small, non-glycosylated proteins.**

How it works

- The gene is cloned into a plasmid under the control of a strong promoter (e.g., T7).
- Upon induction, the bacteria express large amounts of the target protein.
- The product may accumulate in the cytoplasm or in inclusion bodies (aggregates), requiring solubilization and refolding, or even secreted in the case of *Bacillus* or other related host organisms.

Common species

- *E. coli*. The most widely used chassis in synthetic biology
- *Bacillus subtilis* and other *Bacillus species*. Gram-positive bacteria with secretion properties
- *Corynebacterium glutamicum*. Gram-positive bacteria used for the production of amino acids, peptides/proteins, and dsRNA

Advantages

- Rapid growth and low-cost production
- Easily engineered and scaled
- High yield for simple peptides or proteins

Limitations

- Lacks machinery for eukaryotic post-translational modifications
- Product may require complex downstream processing
- Risk of endotoxin contamination must be anticipated (e.g. with some *E. coli* strains) to avoid issues later on during regulatory

Filamentous Fungi-Based Expression Systems

How it works

- Cloning of the gene-of-interest and generation of a production strain is similar to the process as described for yeasts.
- For the production of heterologous proteins/peptides, filamentous fungi are grown in submerged liquid fermentation. Otherwise, they also grow excellently in solid-state fermentation.

Common species

- *Aspergillus niger*. Commonly used to produce enzymes for food industry
- *Trichoderma reesei*. Commonly used to produce industrial enzymes

Advantages

- High secretion capacity for heterologous proteins/peptides
- Good protein-folding capacity
- GRAS status for some species
  Strong promoters and many strain improvement tools available

Limitations

- Filamentous fungi secret many endogenous proteins and so products may require complex downstream processing

Cell-Free Systems (Especially for RNA or Rapid Prototyping)

Cell-free systems bypass the need for living organisms by using **cell extracts** (e.g., from E. coli or wheat germ) that retain the transcription and translation machinery.

How it works

- A DNA or RNA template is added to a reaction containing ribosomes, tRNAs, amino acids, enzymes, and cofactors.
- The target protein or RNA is synthesized in vitro without living cells.
- In the case of RNAi or double-stranded RNA (dsRNA), enzymes like T7 RNA polymerase are used to transcribe long RNA strands from a DNA template, sometimes followed by annealing into dsRNA.

Advantages

- Extremely fast — expression within hours
- No cell-viability concerns (ideal for toxic proteins)
  Ideal for testing libraries or variants in early development
- Flexible for automated or high-throughput workflows

Limitations

- More expensive than whole-cell systems
- Lower yield and scalability challenges
- Purity and reproducibility can vary by extract source

Other, less-established biomanufacturing systems include:

Molecular farming

Molecular farming uses plants for the heterologous production of protein- or peptide-based biopesticides. This approach is well established for pharmaceutical proteins and for high-value proteins with industrial applications.

How it works
- Plants are used to transiently (through transfection) or stably (through transformation) overproduce the peptide or protein of interest.
- Plants are usually grown in greenhouses or under indoor aeroponics/ fogponics conditions.
- The target protein/peptide is then purified from the leaf biomass.

Common species
- Tobacco, mostly *Nicotiana benthamiana*
- Lettuce
- Different algae species

Advantages
- High-level accumulation of proteins is possible
- Some post-translational modifications are possible

Limitations
- Not yet established for agricultural applications due to high COGS and limitations in scalability. Latest innovations such as fogponics that increase efficacy and yield, may change this situation.

Production in Field-Grown Plants

Certain plant species (of the family Violaceae) naturally produce cyclic peptides (cyclotides) with insecticidal properties.

How it works
- The plants are farmed in the field for commercial production.
- Harvesting and DSP conditions are like those of other plant extract-based products.

Species
- Violaceae, in particular *Clitoria ternatea*

Advantages
- No genetically modified organism system involved
- Low-cost cultivation/biomass production
- Cyclotides are very stable which facilitates harvesting, DSP, and formulation

Limitations
- “Crops” are usually not well bred, i.e. agronomic performance (yield) is limited
- This production system is limited to endogenous biomolecules

Solid-State Fermentation (SSF) of microbes (instead of submerged liquid fermentation). To date, SSF is not yet used for the manufacturing of protein- or peptide-based biopesticides. Submerged liquid fermentation is the dominant fermentation type. This is mostly due to a lack of process control, product quality, and scalability in SSF. Recent innovations such as hydrochar-based SSF, have the potential to overcome these limitations. Advantages include extremely low COGS and high titers of microbes grown in SSF compared with submerged liquid fermentation. Prominent challenges include the control of sporulation and fundamentally different DSP steps.

Downstream Process Development

After fermentation (whether done in yeast, bacteria, or cell free), the resulting production material should then go through different purification steps to reach the desired commercial form. This form can be dried or as a liquid concentrate as two examples.

The number and type of DSP steps can vary greatly depending on the production strategy. Examples of downstream steps include:

- Cell lysis
- Centrifugation
- Filtration steps
- Drying

During Phase I, the R&D team should develop the first acceptable DSP to produce the desired form of the TGAI. This part of the process will then be optimized and refined during the process scale-up in Phase II before being frozen for regulatory submission.

The DSP phase often contributes a significant portion of the final production cost, so learning more early can prevent costly business decisions in later stages.

1.3.2 Efficacy Validation of the TGAI

Once a TGAI is obtained with the desired purity and forecasted cost of goods, the next critical step is to confirm that the bioproduced version of the selected lead(s) are still biologically active. While the purity level may be acceptable, the TGAI may still contain various impurities (e.g., salts, lipids), which may have an impact on both the future formulation and efficacy.

During this stage, comparative bioassays should be conducted to benchmark the efficacy of the TGAI version (bioproduced) with the synthetic lab-purified version of the same lead biomolecule. Experiments conducted during step 1.2 (hit-to-lead) are reproduced, but this time adding a modality with a bioproduced sample. The results should give confidence that the biomanufacturing process selected will yield a highly efficacious product.

1.3.3 Stability and Penetration Studies

In parallel to the efficacy validation of the TGAI, the stability and penetrability properties of the lead molecule should be evaluated. Unlike small chemical molecules, **peptides, proteins, and oligonucleotides** can face significant hurdles when applied to plants or pests in open-field conditions. Their larger size and susceptibility to enzymatic degradation present critical **formulation and delivery** challenges. This step of Phase I focuses on evaluating **biological stability** and **tissue penetration** — both of which are critical to downstream efficacy.

Environmental and Biological Stability

One of the main advantages of biomolecules compared with microorganisms is that they generally have a much better stability profile. Unlike microbes, which are living organisms, biomolecules are just simply large molecules; they do not need to remain alive and viable from the manufacturing plant to the farmers' fields.

Biomolecules can have very diverse physicochemical properties: hydrophobic/hydrophilic; positively charged/neutral, etc. Biomolecules can be sensitive to ultraviolet light, pH variability, and to some extent temperature. **In addition, a unique vulnerability comes from biological degradation from proteases, peptidases, DNAses, and RNAses naturally present on and in plants, rhizosphere, and insect guts.** Let's consider the biostability of a few product class examples:

Peptides and Proteins

- Can be degraded by **extracellular proteases** found on leaves, in soil microbes, and inside target organisms.
- A lack of post-translational modifications (e.g., glycosylation or disulfide bonds) can lead to **structural instability**.
- Many peptides are inherently unstructured and can be prone to aggregation or denaturation at non-physiological pH or temperatures.

Olignonucleotide Molecules

- ssRNAs are especially labile; they degrade quickly due to ubiquitous **RNases** in the environment and in pests.
- Contrastingly, dsRNA, used in RNAi technologies, is more stable but still susceptible to:
    - Enzymatic cleavage (e.g., dsRNases in the saliva or gut of insects)
    - UV and oxidative degradation
    - Acidic conditions in the phyllosphere or soil

During this phase, the best lead candidates are evaluated for their biological stability, which is critical for the long-term product efficacy and performance. Peptide, protein, or oligonucleotide stability is monitored over time using high-performance liquid chromatography and mass spectrometry in the presence and absence of various peptidases/proteases or RNases. Biological stability can vary from seconds to hours to several days. The best biomolecule leads with high biological stability are selected for the next phase.

Tissue Penetration: Getting to the Target

Permeability can be a challenge. Depending on the localization of the targeted protein/nucleic acid and the type of product (e.g., curative vs. preventative), the active biomolecule may have to penetrate through the different layers of the plant or insect to reach its site of action. This can be within plant cells, insect gut cells, or even pathogen tissues. The biological membranes that these biomolecules must traverse are poorly permeable to large, charged, or hydrophilic molecules. Depending on the target of choice, the biomolecule may have to be permeable all the way into the nucleus of the cell.

However, because plant cells naturally contain billions of proteins, peptides, and oligonucleotides, the detection of the delivered biomolecule into the plant tissues and cells of interest remains a challenge. Isotope labeling and other techniques are increasingly able to measure how much TGAI entered the cell. However, binary read outs of delivery are significantly easier than quantifying successful delivery. We can consider a couple of examples of permeability challenges to further highlight the importance of penetration:

Plant Penetration

Peptides and proteins can face poor uptake into the plant cells owing to:

- **Cuticle barrier.** A waxy, hydrophobic outer layer of leaves that repels water-soluble molecules.
- **Cell wall and plasma membrane**. Even after breaching the cuticle, biomolecules must traverse tightly structured carbohydrate/protein matrices.

Permeability properties can vary depending on the length of the peptide or protein molecule, its hydrophilicity/hydrophobicity, its charge, or even its amino-acid content. While it has been demonstrated that small peptide sequences or circular peptides could penetrate plant cells and even reach the nucleus (e.g., Green Fluorescent Protein split experiment), there is currently no systematic approach available to rapidly detect, quantify, and compare the penetration properties of different peptide or protein sequences without modifying their natural structure (e.g., adding fluorescent tags would change the physicochemical properties of the natural biomolecule).

Similarly, double stranded oligonucleotides (DNA, RNA) show poor uptake unless they are relatively small in size and **delivered in formulations designed to overcome translaminar and cell-membrane uptake barriers**. Without proper formulation, dsRNA or dsDNA uptake in plants is highly inefficient and frequently limited to natural openings such as stomata or damaged areas on the leaf surface.

Insect and Pathogen Penetration

For insecticidal application, biomolecules must survive in the insect gut. In the case of RNAi, depending on where the target is located, the molecule may have to be absorbed by **midgut epithelial cells** to trigger gene silencing — a process highly dependent on:

- pH of the gut, which differs between insect orders (e.g., Lepidoptera vs. Coleoptera)
- Gut nuclease activity
- Presence or absence of **dsRNA transporters** (e.g., SID-1-like proteins)

Some insect orders (e.g., Lepidoptera) are **inherently poor responders** to environmental RNAi due to low dsRNA uptake or rapid degradation.

During this stage of the R&D process, the main objective is to better understand the intrinsic penetrability properties of the lead molecule selected. This can either be done directly by measuring and quantifying penetrability (although this remains highly challenging from a technical perspective) or indirectly through the combination of MoA confirmation and phenotypic evidence.

Formulation science can also strongly improve the stability and/or penetrability of a given lead biomolecule. During Phase I, the main goal is to find the best active compound that combines efficacy, production feasibility, stability, and bioavailability before any improvement with formulation technologies. Improvement through formulation will, however, be a critical aspect during the Phase II: early product development.

1.3.4 Lead Sequence Optimization and MoA Validation (Optional)

If the results of any of the previous evaluation tests (production feasibility, efficacy of bioproduced material, stability, or bioavailability) are not positive, the next step could either be to change and adjust the design of the lead molecule, focus on another candidate, or simply stop the project.

Peptides, proteins and oligonucleotides can indeed be optimized by adjusting or changing some of the building blocks of the original sequence (e.g., amino acid or nucleotide). The goal is to at least maintain the efficacy level of the original sequence while improving producibility, stability, and/or penetration. During this stage, experiments from step 1.3 are conducted again with each of the variants to find the best optimized sequence to be promoted to Phase II (early product development).

MoA validation

For target-based biomolecule design, biochemicals assays conducted during step 1.1 (design-to-hit) should confirm the MoA of the molecule.

For nature-derived compounds, conducting specific MoA studies could be strategically relevant at the end of Phase I to better understand the product and its relationship to its target, which could support both intellectual property (IP) and future regulatory consideration. Nevertheless, the R&D team should determine “good enough” in terms of MoA understanding.

Phase I Summary: Comparison with Small Molecules

Similarities	Differences
• Target selection and rationale-based design for de-novo design of biomolecule compounds (e.g., RNAi, antibodies, protein interaction) • In-vitro and in-planta screening approaches (efficacy test) • Sequence optimization	• Bioprocess feasibility: “process is the product” • In-vitro to in-vivo translation: penetration can be a challenge • Different early-stage approaches for nature-derived compounds (e.g., protein hydrolysate)

Phase I Summary: Similarities and Differences with Small Molecules

Aspect	CP biomolecules	CP small molecules	Comments
Discovery approach	Target-based or nature-sourced	Mostly target-based	More opportunities for biomolecules
Optimization	Possible	Possible	Biomolecules: often for improved efficacy and lower COGS Small molecules: often for environmental safety
Production	Biomanufacturing often required	Chemically synthesized	Chemical synthesis is often cheaper and easier to scale; biomanufacturing is more sustainable
Molecule size	1kDa to >150kDa	<1 kDa	Smaller molecules are often easier to formulate and have longer shelf life
Penetration challenge	High; need delivery systems	Low; penetrate easily	This limits the efficacy of biomolecules, but is also a cause for the safety concerns of chemicals
Cost drivers	Bioproduction	Synthesis, tox/ecotox testing	Cost drivers for biomolecules are foreseeable, those for chemicals are not

Phase II:
Early Product Development

Timing

18 to 36 months

Summary

Phase II marks the transition from research to development.

The two main goals of Phase II complement each other: (1) establish and scale a manufacturing system that can be commercially viable and use it to produce enough TGAI to (2) perform formulation studies and field trials to allow reliable estimates of efficacy, pest spectrum, and other product-related parameters

Main steps during Phase II for biomolecules:

- Step 2.1: lead optimization and early field trials
- Step 2.2: scale-up feasibility of the manufacturing process
- Step 2.3: develop formulation prototypes
- Step 2.4: check FTO, file IP, prepare for regulatory

Cost Range & Trend

~$4 to $9million

Remaining neutral. Establishing a manufacturing system that yields sufficient amounts and quality of TGAI for meaningful formulation and field-testing studies can vary according to biomolecule product types. However, synthesis methods continue to improve

Opportunities & Common Pitfalls

Opportunities

- Proper experimental design at each level of greenhouse, semi-field, and field trials to efficiently test the multiple variables (e.g., use rate, timing of application, different formulation) impacting product positioning and maximize the learnings from these trials
- Set up efficient project management office (PMO) to efficiently coordinate the necessary work across multiple areas of expertise including but not limited to chemistry, formulations, biomanufacturing, biology, and data science; ensuring the delivery of results on time before the next season's trials need to be deployed
- Access to proper production scale-up facilities (contract development manufacturing organization; CDMO) to rapidly derisk the scalability of both the production steps (USP) and the purification steps (DSP)
- Performing early toxicology studies to rapidly identify potential red flags before too much money is spent on a poor candidate

Common Pitfalls

- Improper field trial design that lacks statistical significance or positive controls. There are only two opportunities per year (including counter-season) to learn from the field.
- Choosing commercially irrelevant crops and/or pest(s)
- Failure to identify the regulatory path, as well as the long-term costs and time to market
- Focusing only on USP improvement without considering impact on DSP steps
- Overestimating process scalability: conditions are not the same in 1L vs. 1000L bioreactors. Failing to meet target cost for a commercially viable product

2.1 Lead Optimization and Early Field Trials

Lead optimization and early field trials in Phase II are crucial to take AI-based R&D to TGAI-based R&D. Specificity, selectivity, and stability data obtained with pure AI need to be confirmed at TGAI level.

- Lead-to-TGAI transition: Successful transition from pure compound (e.g., peptides made by chemical synthesis or peptides/proteins produced in a research-stage, inefficient bio-manufacturing system) to the TGAI, which is the “real” bio-produced material containing the AI at a given purity level. The TGAI should be produced in a close-to-commercial manufacturing system and be at the purity level of a potential commercial product.
- Lab work includes:
    - Basic stability studies of the TGAI over time at different pH values, temperatures, and osmolarities/ionic strengths.
    - In Phase II, the evaluation of selectivity and spectrum of activities against target pests as well as the absence of toxicity towards non-target organisms (in-vitro/in-vivo work) must be confirmed on a TGAI level and will be further expanded.
    - It is important to choose bioassays that are relevant and translate to the field (e.g., for bioinsecticides with the target in the midgut, ingestion assays (and not contact assays).
- Greenhouse work includes:
    - Preliminary formulation testing with the TGAI is important to identify the specific formulation needs of the product candidate (sticker, penetration, stabilizer).
    - Understanding the dose-response and improving the use rate. Both are pest, pathogen, and crop specific. Understanding these parameters for key pests/pathogens and crops in the greenhouse is key for the planning of meaningful field trials.

Measuring the Impact of the Timing and Number of Applications

Does the biomolecule have a curative effect, or is it limited to preventative applications? This is of particular importance for biofungicides/bacteriocides. How long does the biomolecule stay active after spraying? Answering these questions will help to determine the timing of application in the field.

Greenhouse-to-field translation:

- Semi-field trials are a good way to bridge the gap between controlled lab/greenhouse studies and open-field trials. Usually, plants are in pots but outside in the real environment, often using net houses that resemble outdoor light, pest, and pathogen conditions.
- Inoculated trials (i.e., trials with artificially created pest pressure) are crucial to better understand the performance of the product candidate under different pest/disease pressures.

- Small-scale field trials should be performed as soon as a TGAI formulation is available. Only field trial results provide the final evidence that the AI works in the field and may become a commercial product. Usually, start-up companies go through a steep learning curve regarding field trails. Therefore, the first-year field trials should be performed early, and expectations should be low. Some recommendations for small-scale field trials:
    - They should be close to your site (so they can be monitored frequently)
    - Location and timing must ensure a good disease/pest pressure
    - Since disease/pest pressure can never be guaranteed, trials in multiple locations will reduce the risk of low disease/pest pressure
    - A positive control (chemical pesticide) must be included; as should a commercial biopesticide, if applicable
    - Key species of the most important crop groups (relevant for Environmental Protection Agency [EPA] registration) should be used
- The objectives of small-scale field trials are:
    - To obtain a first proof of concept of the TGAI/AI efficacy potential in the field,
    - To start comparing your product with (chemical and biological) products in the market
- Small-scale field trials will usually take one or two seasons. Theoretically, one season is sufficient. The use of Southern and Northern hemispheres allows doubling of field trials within one season. In reality, a start-up company's small-scale field trials of a first product will require two seasons.

2.2 Scale-Up Feasibility of the Manufacturing Process

A key goal of Phase II is to establish and scale a manufacturing system that is likely to be commercially viable. This is a prerequisite for being able to perform formulation and field trial studies. It provides the basis for a reality check on manufacturing costs and COGS.

For microbial strain-based manufacturing, scaling of manufacturing includes the development of a stable, commercial production strain with high-level production of the target protein/peptide.

Scaling of manufacturing in Phase II is usually from one or two liters to hundreds or thousands of liters. At the beginning of Phase II, this may involve the comparison of two different production systems. A decision on the preferred production system, however, should be made as early as possible. Many parameters can change during scaling:

- Steel fermenters with inline analytics; seed train
- Cheaper media components
- Oxygenation, shearing and feeding parameters
- Fed batch vs. continuous fermentation
- Use of DSP equipment similar to that in commercial scale manufacturing (20,000-100,000 liters)

In the case of dry formulations, adequate drying steps (spray dryer, fluid bed dryer) are included. Choices depend on the heat stability of the product and the desired target formulations (e.g., WDG). This process scale-up involves working with CDMOs. The right choice is important; factors to improve the chances of this, include:

- Experience with the production system and commercial strains
- The ability to perform large-scale manufacturing, if possible, as this eliminates the extra step from pilot-scale manufacturing CDMO to large-scale contract manufacturing organization (CMO)

A preliminary manufacturing process map should be developed — as a reference for further modifications and improvements and as basis for a preliminary COGS analysis based on fermentation and DSP performance.

Analyses include the quantification of the product, and identification of sub/by-products. Fast and relevant bioassays should be performed using samples from key steps of the manufacturing process.

2.3 Develop Formulation Prototypes

It is key to identify (and be able to commercially produce) the ideal formulation of a biomolecule pesticide. Each potential formulation has pros and cons, and not every formulation will be suitable for every AI.

Liquid concentrates (LCs), for example, have the advantage of reduced DSP, since there is no drying step involved. On the other hand, LCs are problematic for many AIs because of instability at a TGAI level. LC formulations are also more prone to contamination, usually requiring the addition of antimicrobial compounds (e.g. Methylisothiazolinone, Benzisothiazolinone), **and** may have limited shelf lives compared with dry formulations.

Dry formulations obviously require a drying step, which needs to be carefully chosen based on the stability and heat sensitivity of the AI. Standard spray drying (with temperatures >100°C) is often not possible for proteins, peptides, and RNA-based products. Adjustment of spray-drying conditions and specific spray-drying variants allow use in heat-sensitive AIs. Outlet temperatures can be reduced, and electrostatic spray drying has been validated for many protein-based actives. This allows simultaneous drying and encapsulation of the AI. Another gentle drying technique is fluid bed drying where the evaporative cooling effect helps maintain a relatively low and uniform temperature throughout the drying process. New techniques such as rapid room-temperature aerosol dehydration have only been used to date for pharmaceutical proteins but have potential for broader applications including agriculture.

The most important dry formulations for protein-/peptide-/RNA-based biopesticides are wettable powders and WDGs, the latter being preferred by farmers for safety and ease of handling.

Formulations may also include encapsulation of the AI to improve its stability, penetrability, tank-mix compatibility etc.

In Phase II, the formulation needs and options for the AI must be identified, and promising formulations advanced to a stage that can be scaled. It is important to have formulation expertise and a small formulation lab/team in house. Certain approaches (e.g., encapsulation), however, may require collaborations or partnerships with dedicated formulation technology service providers.

A comparison of the properties of the TGAI vs. the pure AI can often be performed in house. This includes:

- Determining the physicochemical properties (e.g., solubility). This is important for tank-mix compatibility
- Performing stability tests at a TGAI level to test:
    - Potential degradation at different pH and temperature conditions Accelerated shelf-life studies. Is the TGAI suitable for standard conditions (e.g., 54°C for 14 days)? This would shorten development timelines.
    - UV-light stability, which is key for all field applications. UV sensitivity can often be overcome with encapsulations
- Conducting penetration tests, e.g., formulations that allow the AI to cross the plant cuticle and cellular membranes
- Performing rainfastness testing

The development and testing of formulations in the lab, greenhouse, and field is a complex task and requires a small cross-functional team with broad expertise (biochemistry, chemistry, biology, agronomy). Formulation studies also involve mixture testing and multiple compatibility tests:

- The TGAI is tested in different buffers, in combination with adjuvants, stickers, spreaders, penetration aids, etc
- Test compatibility with other AIs/products (tank-mix compatibility)
- Tank mix with chemicals (other pesticides) and with fertilizers (respective salt solutions and pH may lead to precipitation of protein-/RNA-based AIs)
- Compatibility will ensure acceptance by the farmer and eventually commercial success for the company.

Beyond the classical formulation toolbox, a variety of encapsulation technologies have been developed for (bio)pesticides to improve solubility, tank-mix compatibility, (UV) stability, penetrability, and rainfastness of biomolecules. Collaborations/partnerships with dedicated formulation technology service providers can enable the testing and implementation of specific biomolecules.

Comparison of encapsulation technologies for biomolecule biopesticides:

Technology	Applications	Pros	Cons	Examples
Liposomes	All biomolecules	Relatively low cost	Wide range of vesicle sizes	
Lipid nanoparticles	All biomolecules	Proven in pharma	Rel. high costs; no commercial products in ag	
Polymeric nanoparticles	All biomolecules	Commercial formulations available; compatibility with fertilizers;		Most important: chitosan nanoparticles; others include PEG-based polymers
Fermentation-derived encapsulation	All biomolecules	Fermentation and formulation can be combined		Several products in development
Bioclay (layered double hydroxide)	dsRNA products	Low cost	Limited encapsulation efficacy/ yield	Products in development
Virus-like particles	dsRNA products	Reduced risk of insertional mutagenesis	Limits manufacturing to E. coli	
Carbon dots	dsRNA products	Low cost, effectively increases half-life of the AI, facilitates transmembrane delivery		RNAi platform (in plants), products under development

2.4 Regulatory, Safety, FTO, and IP

Intellectual Property

Establishing a strong IP portfolio is a key task for any venture capital-funded start-up. This starts in Phase I with a freedom-to-operate (FTO) analysis and potentially execution of necessary licensing steps. The filing for a patent protecting a a biomolecule or (ideally) an entire class of biomolecules ideally with a novel MoA begins in Phase I as part of the development (and execution) of an IP strategy.

In Phase II, this patent portfolio can be expanded by new patent filings on novel formulations (e.g., encapsulations), uses, and manufacturing processes. Alternatively, select IP such as optimized manufacturing processes can be kept as a trade secret. Patent filing can enable marketing and commercialization strategies and is often expected by investors and potential acquirors, but it is expensive and involves publication sufficient for others to make and use the invention without knowing if a patent will be granted. Publication may also allow competitors to develop workarounds and improvements. IP creation on novel formulations, uses, and manufacturing usually begins in Phase II and continues into Phase III.

Safety

A key advantage of biomolecules compared with chemicals is their safety profile (for humans, beneficials, and the environment). Certain safety aspects can be explored in Phase I. These include basic toxicity tests comparing activity against target organisms vs. non-targets (e.g., pest vs. beneficial; weed vs. crop) or in-silico allergenicity testing (using sequence alignments and epitope mapping).

In Phase II, in some geographies, a broader range of preliminary safety studies will be performed with the AI and TGAI. These include genotoxicity testing using a micronucleus test, the Ames test to exclude carcinogenicity, and testing the AI against beneficial organisms (e.g., honeybees, lady beetles, lacewings, predatory wasps, earthworms, fish). All mentioned toxicity studies are relatively standard and thus can be outsourced to respective regulatory service companies. Environmental fate studies are usually straightforward for biomolecules since the AIs break down into short peptides, amino acids or nucleotides, which are of little concern from an environmental fate perspective.

Several safety studies need to be performed with the TGAI and formulated product. It must be ensured that the production host (in case of fermentation-based manufacturing) does not contribute any harmful compounds to the product. The use of a production host with GRAS status is therefore recommended. The same applies to formulation components. All inerts (often added to allow an efficient drying process) and encapsulation materials must be reviewed and (if needed) tested for their safety profile. The TGAI and the formulated product also need to be checked for any phytotoxicity at relevant developmental stages of target crops.

Regulatory

In Phase III we will discuss the work packages needed to generate a regulatory data package. To do so, a regulatory expert should be engaged (or employed) in Phase II. This will ensure early and professional engagement with the regulatory bodies in the target market. For registration in the US, this would be the Emerging Technologies Branch of the Biopesticides and Pollution Prevention Division within the EPA's Office of Pesticide Program. This involves the application for a biomolecule classification at the EPA.

A data gap analysis should be performed in Phase II to inform a regulatory strategy including timelines and costs. For the registration of biomolecule pesticides, the most expensive and time-consuming parts are chronic toxicology guideline studies (oral, dermal, inhalation, and prenatal development; each study takes more than 90 days with total costs of around $1 million) as well as efficacy trials (usually for several crop groups and pests/pathogens; performed at multiple locations, the total costs are $300,000–500,000). These studies should be planned and budgeted in Phase II. Contract research organizations (CROs) for guideline studies and efficacy trials should also be selected and engaged.

Phase II Summary: Similarities and Differences with Small Molecules

Similarities during Phase II:	Differences during Phase II:
• Efficacy assays • Evaluation of spectrum of activity • Field trial scale up and testing	• Importance of biomanufacturing process development and transition from AI to TGAI • The importance of encapsulation and formulation technologies for reaching the target of interest and ensuring good stability

Phase III – Phase V: Development

Phase III
Advanced Product Development

Timing

24 to 36 months

Summary

Phase III, much like in crop-protection small molecules, focuses on the scaling up of testing in the field to better define the product parameters for use. Efficacy, dosing, utilization, and pricing of production are all better understood by the end of this stage.

This phase also focuses on scaling up production capabilities and preparing the five regulatory batch requirements by finalizing and locking in the process ("process freeze"), TGAI, and at least a first formulation.

This Phase includes the following efforts:

- Efficacy testing
- Production development, process freeze, and five-batch study
- Formulation preparation and formulation freeze
- Cross-resistance studies
- Regulatory preparation, safety, FTO, and IP

Opportunities & Common Pitfalls

Opportunities

- Same as during Phase II, excellent Project Management would help streamline all required studies and maximize learnings at each step. Having in-house regulatory expertise is a must-have at this stage.
- Engaging early with regulatory agencies whenever possible to confirm regulatory strategy and data requirements. Ask for waivers whenever possible to simplify the regulatory work.
- Begin to explore go-to-market partnerships and joint development trials in the field to access broader testing and commercialization opportunities.

Common Pitfalls

- Process development: freezing the "wrong" process or changing the process book mid-way. Remember, regulatory work cannot start before having locked in the production process. And once it starts, changing the process would mean restarting all the regulatory work.
- Assessing toxicological risks too late: toxicity studies should be performed early (sometime in Phase II) to avoid nasty surprises later.
- Not or partially interpretable field trial results due to incomplete experimental design, poor preparation, etc. Each season in the field must bring the company a step closer to identifying the best value proposition for the farmers who will use the future product.

Cost Range & Trend

$7 to 10 million

3.1 Efficacy Testing

Efficacy is the cornerstone of any crop-protection product. In this phase the efficacy of biomolecules is better understood and standardized through scaling of the field-testing program. These field trials should happen across the crops and geographies of interest and compare against the national, regional, and local best-in-class products as benchmarks (positive controls).

During the multi-year and multi-locations field-trials campaigns, the technical team should try to best understand how to position the future product to provide the best value for the growers. Key parameters tested in the field during this phase typically include:

- Efficacy vs. existing products and programs (positive controls), especially comparing with existing commercial biological references
- Performance of the product alone (solo) vs. performance when used in-combination with other products (tank-mixed, co-packaged, or in an application program with small-molecule products). Although solo performance is a must, demonstrating that the future product could easily be combined with other would be a plus.
- Application rate and dose-response studies: refined each year from previous experiments, and potentially adjusted to specific crops or pest targets (effective use rate could vary depending on the crops and/or pest target)
- Formulation exploration, refining and selection to be in a position to “freeze” a first formulation for regulatory studies.
- Yield quantification and economic analysis: comparing the ROI for the growers of various programs to demonstrate the real value proposition of the future product

As mentioned before, proving the efficacy of the biomolecule products in the best field conditions can be challenging.

- Biomolecules products generally have different MoAs than chemicals and may not readily penetrate in their target organisms.
- While chemicals mostly target internal cellular pathways that are mobilized during the entire lifecycle of the target organisms, biomolecules can have a more specific target that usually requires the product to be sprayed at a more accurate development time than chemicals.
- Biomolecules are more sensitive to the environment than chemicals. While this can be addressed by adequate formulation, it is important to consider when comparing with certain long-lasting effects of chemical products.

At the end of this phase, the company should have a comprehensive understanding with a significantly relevant set of data to demonstrate the superiority of the product in specific conditions, within specific programs against existing commercial products.

3.2 Production Development, Process Freeze, and Five-Batch Study

It is rare that the company has internal resources and capacity to fully develop the production process internally and will most likely rely on **production development** partners or CDMOs. At this stage, the process book is being developed together with the CDMO partner, and the techno-economical model is fully established to identify the unit cost in function of the production volume, COGS, and the application rate.

If the company is developing a brand-new biomolecule that has never been scaled up for agriculture, the process is likely to be an order of magnitude more challenging; followers have much less risk during this phase if they can lean on existing processes and avoid pioneers' pitfalls.
The most impactful decision during this phase is probably the "**process freeze**": the company should lock what it believes to be the best production system before any regulatory work starts. Once the process is frozen and the regulatory studies start, the company will have very few options to change it without restarting all registration studies.

Once the process is frozen, the company should demonstrate its reliability by performing a **five-batch study**, preferably using the facilities of the future commercial production site. During this study, the company should produce five different batches, analyze each of them, and demonstrate that the quality and purity of the material produced always stay within the acceptable range. For biomolecules, these specifications should be flexible enough to accommodate for the variability of the biological processes required for manufacturing. A TGAI with 99.9% pure AI is not required, as long as the other components (impurities) in the TGAI are clearly identified and obviously not problematic. Large standard deviations in the regulatory dossiers allow for fermentation batch variation and will save the company the challenge of requesting future modification in the regulatory dossier that can delay time to market. In Europe, the acceptable range is typically three times the standard deviation in terms of purity (%).

Performing the five-batch study establishes a quality control baseline that ensures consistency and product quality of future production batches. The five-batch study is a critical milestone in the product development journey and usually signals the start of regulatory studies, which have to be performed using the TGAI from one of these five batches.

For products that do not require fermentation for production, such as using chemical synthesis or extraction of natural biomolecules, the requirements will be comparable to those for chemical AIs. However, similar requirements when it comes to process freeze, production partner identification, etc. will be required at this stage when submitting for regulatory approval.

3.3 Formulation Preparation

In parallel to production development and process freeze, the company should decide and lock in a first formulation ("**formulation freeze**") for the regulatory studies. Formulation is likely to be refined during Phase III, pending field trials studies. The aim could be to improve efficacy, product shelf life, or duration of effect in the field. Alternatively, it could be optimized for marketing, including certifications for the product to be used in the most adapted environment (organic farming, indoor, etc.).
As with the production process, changing formulation midway through the regulatory studies could delay regulatory approval and complicate the registration process. However, once the TGAI and the first formulation are approved, it is usually cheaper and faster to obtain another authorization for a second formulation (as long as the TGAI is the same). As the company continues

to improve formulation, it should wait for the first formulation to be approved before submitting another one, unless it is a complete game changer.

3.4. Cross-Resistance Studies

Another type of study that could be performed during Phase III and which could support the product positioning is to demonstrate absence of cross-resistance against key pathogens with existing chemical AIs.

This study may serve as a marketing and competitive positioning tool for market entry.

3.5 Regulatory Preparation, Safety, and FTO

During Phase III, the focus should be on preparation for submission to the regulating bodies in the geographies of interest. The company should refine its understanding of the required data packages, find appropriate lab partners and collaborators (CROs) to perform the studies, and estimate the costs of regulatory studies and approval.

Key regulatory activities during Phase III include:

- Engaging with all appropriate regulatory entities in each target country, either directly or through a network of regulatory consulting agencies.
- Requesting waivers. Most biomolecules are readily biodegradable, which means they will not accumulate in the environment and instead degrade to their building blocks (amino acids, bases of DNA/RNA). Therefore, if supported by strong scientific evidence, certain mandatory regional regulatory requirements may be waived by the authorities at the company's request.
- Engaging qualified CROs to perform key studies:
    - Analytical analyses
    - Toxicity studies (human, beneficial insects, etc.)
    - Eco/aquatoxicity studies (soil and water microorganisms)
    - Physicochemical studies (e.g., degradation – shelf or on-seed stability may be required)
- Whenever possible, conduct "de-risking" studies with future CROs. This is not mandatory at this stage but could significantly facilitate the "real" regulatory studies in Phase IV. For example, calibration studies for the analytical studies usually require multiple back-and-forth iterations between the company and the CRO.

Planning for regulatory submission in different regions

The company may elect to prioritize one or more regions for the first market entry of the product. Certain regions tend to review dossiers more quickly (Brazil, USA) while others may take more time and may not have a dedicated regulatory path for biomolecule crop protection products (EU). While the cost is multiplied by the number of regulatory demands, certain costs may be saved as studies can be used in multiple dossiers at the same time.

For example, the regulatory path in the US and Brazil is usually simpler for biological products, with dedicated and defined paths (biomolecules, microbials, or new technologies). They propose data requirements specific to the type of technology submitted with the potential to accelerate the overall regulatory process. In Europe, the approval process for agricultural input products can take significantly longer because multiple regulatory bodies are involved. These include the Rapporteur Member State, EU-level authorities such as EFSA, and each individual member state that must grant its own approval. All products must follow the same regulatory framework, Regulation 1107/2009, which adds complexity and time to the process. These regulations are continuously evolving, and regulatory experts should be consulted.

Pertinently, however, the company can also request an EUP (emergency use permit). This permit allows the product to be tested on a larger, commercial scale without requiring the crop to be destroyed afterward, which is a standard part of many experimental field trials.

The decision to pursue regulatory approval with one or multiple agencies at this stage will depend on the financial and human resources of the company as well as potential support obtained from commercial partners already in play at this stage.

Phase III Summary: Similarities and Differences with Small Molecules

Similarities during Phase III:	Differences during Phase III:

• Scaling up field trials to establish efficacy vs. best-in-class positive controls and refining product positioning
• Preparation for regulatory approval
	• Production scale-up – specific to the bioprocess technology followed
	• Regulatory studies: Expected to be lighter, faster, and cheaper than with crop-protection small molecules (based on current approvals). For biomolecules these studies either:
	  • Basic stability studies of the TGAI over time at different pH values, temperatures, and osmolarities/ionic strengths.
	  • In Phase II, the evaluation of selectivity and spectrum of activities against target pests as well as the absence of toxicity towards non-target organisms (in-vitro/in-vivo work) must be confirmed on a TGAI level and will be further expanded.
	  • It is important to choose bioassays that are relevant and translate to the field (e.g., for bioinsecticides with the target in the midgut, ingestion assays (and not contact assays).
	• Differences in study preparation for analytical analysis due to the unique nature of biomolecules. Analytical chemists/biologists may need to adjust/develop appropriate analytical methods for this class of products

Phase IV:
Pre-Launch Preparation

Timing

24 to 48 months

Summary

Phase IV moves the product much closer to its commercial realization, and aims to reach the following endpoint:

- **The 'product' is defined**, both TGAI and its formulation
- **Pre-market field trials are complete, and the go-to-market plan is established** with distribution agreements being prepared in relevant geographies
- **The process book is finalized** and transferred from CDMO to CMO; the company has established diversification in its sourcing of the product or intermediate ingredients
- Regulatory dossiers are submitted for review: all studies are completed, and the registration dossiers are filed in each target geography.
- **Product stewardship plan is put in place** including details on technical specifications, label information, quality control, and packaging

The main considerations and steps in Phase IV:

- Field Trials Campaigns: Regulatory and Pre-Market Trials
- Commercial Production and Formulation
- Regulatory Dossier Submission and Support
- Product Stewardship Plan

Opportunities & Common Pitfalls

Opportunities

- Submitting a registration dossier in a country with small market potential but fast regulatory approval as a test launch market. This strategy can help obtain approval in other markets earlier and derisk the future commercial launch in a bigger country with a slower regulatory process
- **Derisk commercialization** by working with growers, extension specialists, and potential commercial partners to pre-book orders for the future product
- Pursuing regulatory waivers to significantly speed up the approval timeline
- Accessing existing and compatible commercial manufacturing lines (CMO) to prepare industrial-scale production, formulation, and shipping without having to build custom facilities

Common Pitfalls

- Tier I regulatory studies with unfavorable outcomes that lead to additional, more expensive, longer, Tier II studies
- Underestimating the length of the review period once dossiers have been submitted (e.g., could be >40 months in Europe)
- Issues with manufacturing and formulating, whether from a technical, quality, or unit-economic perspective
- Failure to engage with growers, key opinion leaders, or potential commercial partners

Cost Range & Trend

$10 to 24million

Depends on the number of crops and geographies of interest

4.1 Field Trials Campaigns: Regulatory and Pre-Market Trials

During this phase, the company should first perform the required regulatory field trials to be able to submit the dossiers. Depending on the country, regulatory agencies usually require either one or two seasons of field trial data to be included in the registration dossier. Regulatory trials should be performed by registered CROs and usually cost more than R&D field trials.

Once the regulatory field trials have been done and the dossier submitted for review, the company will have to wait for approval before being authorized to sell its product. This period could last from anything between 12 to 40+ months depending on the countries. As it waits for approval, the company will prepare its future commercial launch by conducting pre-marketing or pre-sales trials with several partners: key opinion leaders (e.g., universities, influential growers), distributors, industry incumbents, etc. These “validation trials” will prepare commercial traction and support the product claims and unique selling proposition.

The company can also use this period to expand EUP to start testing the product in additional geographies, crops, or pest targets to prepare potential product expansion. Similarly, if the company has enough resources, it can start working on additional formulations, which could be filed after the first product is approved.

4.2 Commercial Production and Formulation

During this phase, if not done already, the company should transfer production from CDMO to the industrial CMO who will oversee commercial production (and if possible, formulation, packaging, and shipping), with a finalized, stable and compliant process.

The company should initiate and validate in multi-10m3 production capacity in real commercial production operations (which can be very different and less flexible than CDMOs). These large-scale production campaigns should:

- Comply with the process book submitted for regulatory approval
- Give results within the specifications (quality, purity, etc.)
- Confirm the techno-economic analysis, cost, and margin projections
- Support regulatory and pre-commercial trials with the same quality consistency of both the TGAI and the final formulated product

In parallel with production, the company should clearly define the product specification for quality control along the production, formulation, packaging, and distribution steps.

If the five-batch study from Phase III was not done with the new CMO, another five-batch study will likely be required to qualify this new production site once the dossier is approved.

Finally, the company can start thinking about diversifying production supply to allow for local production, if possible. This is more sustainable as well as decreasing the risk associated with a single production site. However, each production site would have to be qualified and registered with a specific five-batch study (using the exact same process).

4.3 Regulatory Dossier Submission and Support

During this phase, the company should conduct and finalize all regulatory studies and complete the registration dossiers for submission in each target geography. Gathering all data could take from 12 to 24 months, depending on the number of regulatory trial seasons needed and the complexity of the studies. Regulatory studies should be performed on the TGAI from the five-batch study done at the end of Phase III. After submission, the data will then be reviewed by the appropriate regulatory agencies for each target country.

This review could last 12 to 40+ months. During this reviewing period, the company should continue to engage with regulatory agencies to answer any questions, provide additional data, and prepare the future commercial launch of the product in parallel.

During this phase, the company can also explore additional EUP to start expanding the product claims (additional crops, other pests, etc.).

4.4 Product Stewardship

Product stewardship is akin to that outlined in the chapter on crop-protection small molecules. However, the following guidelines from CropLife provide a great reference resource for background information:

https://croplife.org/wp-content/uploads/2024/12/CLI-Stewardship-IPM-Guideline-2024.pdf)

Phase IV Summary: Similarities and Differences with Small Molecules

Similarities during Phase IV:	**Differences during Phase IV:**
• Conducting pre-market and regulatory field trials for submission • Engaging in proper product stewardship • Developing a go-to-market strategy with key partners	• Production scale-up at the commercial stage: CMOs are more specific for biomolecules and limited Capital expenditure (CAPEX) investment for specific fermentations might differ and increase costs

Phase V:
Launch and Market Expansion

Timing

12 to 24 months

Summary

During this last phase, the company finally obtains authorization to sell its product. During the first few years of commercialization, the company should also look to expand its commercial opportunity by:

- Developing data on adding additional crops and/or geographies to expand its serviceable market size
- Look into developing additional formulations to expand the product label and commercial opportunity

The main considerations and steps in Phase V are:

- Production and Distribution Optimization
- Market, Document, and Engage
- Continuous Product Improvement
- Product Geographic Expansion

Opportunities & Common Pitfalls

Opportunities

- Collaborating with go-to-market partners to rapidly accelerate market reach
- Leveraging early wins as testimonials for product performance
- Streamlining commercial and market development trial costs and allowing that to accelerate the discovery and development of "2nd Generation" product(s)
- Utilizing existing regulatory approvals to accelerate approvals in new geographies

Common Pitfalls

- Underestimating the cost and manpower needed to market and distribute products to farmers and deciding to go it alone
- Refusing to listen and learn from negative market feedback on the product
- Failing to continue to test the product performance across multiple conditions in combination with the latest farm practices, which are always evolving

Cost Range & Trend

$8 to 15million

5.1 Production and Distribution Optimization

Establishing the supply chain with manufacturing, formulators, and packing partners requires different capabilities than used in earlier phases. This step should often be done with industry partners that are also interested in distributing the product. With continuous changes to national and international trade and regulatory policy, diversifying the production of the biomolecule or the other elements of product formulation is critical for long-term viability.

Selling directly to farmers is the dream of many companies, to minimize intermediaries and increase margins. However, with well-structured distribution channels in the major global Ag markets, it is often more effective to leverage existing distribution channels to enable large-scale distribution even if it means decreasing margins. Securing critical distribution and go-to-market partnerships may make it easier for small companies to focus on their core business in product development and innovation.

5.2 Market, Document, and Engage

Even in the case of engaging distribution partners, the company may want to engage in a holistic marketing campaign for the new biomolecule product. Is it critical to pair the marketing campaign materials with a large amount of documentation based on efficacy and safety data that have been validated by the regulatory agencies. These documents contain, but are not limited to, safe use of the product with notion of protection of operators, full product material safety data sheet (MSDS) info, product label and use instructions, and tank mixability. It is the responsibility of the company to ensure proper product safety and labeling information makes its way to the end consumer/farmer.

Engaging with the farmer customer as the product enters the market is critical for accessing key product feedback. Positive feedback can be used as testimonials (when given permission) for additional marketing campaigns. However, negative feedback is also important to further improve the positioning of the product. Like all products, the ability for a company to adapt to negative feedback will contribute to future product improvements and innovations.

5.3 Continuous Product Improvement

In this phase, the company can use customer input to shift from a single TGAI. It can begin to develop a range of different products for application in different crops or increasing the efficacy against a broader range of pests or pathogens. The portfolio strategy includes development of new formulations, development of product mixtures to expand the efficacy spectrum, as well as new packaging size for different market segments, for example. This provides a shortcut through the earlier testing phases for these second-generation products.

With the biomolecule product(s) now registered, the field trial testing now occurs in a full commercial system. This means it is possible to test and trial without the need to destroy the crops post growth, as in earlier phases. Growers, industry partners, and third-party analysts can see product performance in real commercial environments, including with different product rotations for integrated pest management. The efficacy of the product plays an important role to facilitate product adoption in the early phase, but how it works in a farmer's full system is paramount to long-term market success.

5.4 Product Geographic Expansion

After launch in the initial target markets, the company should plan for market expansion. This planning includes engaging with new regulatory agencies, and frequently leveraging the approvals already obtained in other countries/regions. For biomolecule products that have limitations in stability and transportability, identifying options to produce close to the region of commercialization could be quite interesting to generate long-term savings. Identification of new CMOs and expansion of the supply chain is a good step to minimize the risks associated with product availability in different regions.

Phase V Summary: Similarities and Differences with Small Molecules

Similarities during Phase V:	**Differences during Phase V:**
• Go-to-market strategies will be similar depending on the market • Customer and direct farmer engagement to drive belief/trust • Leveraging existing regulatory approvals to accelerate entrance into new markets	• Production considerations associated with shelf-life and transport • Labeling and safety requirements may differ depending on the region

Summary of Crop-Protection Biomolecules

Biomolecules are a critical emerging class of products that offer high specificity and novel MoAs. While these products must overcome key hurdles like delivery to the target or production costs, they can be efficacious and safe products that are highly complementary to current best farm practices.

Cost Summary	Estimated Time to Market
Research **$5 - $13m** **Development** **$25 - $49m**	**Research** **2 - 5 years** **Development** **5 - 9 years**
Total Cost **$30–$62m**	**Total Time** **7–14 years**

Field Testing Guide

Number of trials: depends on the number of different targeted crops and different targeted climates / geographies

Average size of each trial: depends on the number of conditions to test (ex. different formulations, different timing of applications, different dosage, etc.) and on the crop type (ex. trees vs. row crops)

TGAI Samples Quantities: depends on the number of conditions tested, the size of the trials and the dose range per ha

Field Trials Campaign Guidelines - INDICATIVE ONLY

Phases	Years	Main Goals of field trial campaigns	Seasons (indicative)	Indicative number of trials (range)	Indicative Average size per trial (range)	Indicative total trial area (range)	Indicative TGAI Sample Quantities Range	Selection of Key Milestones (illustrative)
Phase 0 Define the problem	1	n.a.	n.a.				n.a.	Techno-Economic Analysis (TEA)
Phase I Pre-field Discovery	1	n.a.	n.a.				n.a.	Leads Selection
Phase II Early Product Development	2	1. Demonstrate efficacy of TGAI in real conditions vs. benchmarks 2. Test different doses, timing of application and intervals 3. Test tank mix compatibility 4. Gather feedback from field users	North Hem.	3 to 5	50-100 m²	150-500 m²	15-50 grams	
			South Hem.	5 to 10	50-100 m²	250-1,000 m²	25-100 grams	Efficacy Proof of Concept in field conditions
	3	1. Confirm efficacy of different production batch 2. Start evaluation of best timing of application, best dose (g/ha) and intervals between treatments 3. Test and compare multiple formulations 4. Demonstrate consistency across soil type and wheather conditions 5. Identify limitations (ex. dosage, timing, conditions, etc.) 6. Start preparing Regulatory Trial plans (including permit)	North Hem.	10 to 20	50-100 m²	500-2,000 m²	50-200 grams	
			South Hem.	10 to 20	50-100 m²	500-2,000 m²	50-200 grams	Positioning in the field and updated TEA with preliminary value proposition.
Phase III Advanced Product Development	4	1. Refine dosage, timing of application and intervals 2. Confirm efficayc of Production batch to validate process freeze decision 3. Freeze first commercial formulation 4. Demonstrate compatibility/synergy with existing programs 5. Quantify economic gain (yield gain and $/acre gain for the farmer) 6. Prepare Regulatory Trial plan for the next year (including permits)	North Hem.	20 to 30	100-200 m²	2,000-6,000 m²	200-600 grams	
			South Hem.	20 to 30	100-200 m²	2,000-6,000 m²	200-600 grams	Process Freeze Formulation Freeze
	5	1. Confirm efficacy and economic gains (yield and $/acre) with TGAI from the 5-batch and the selected formulation 2. Refine dosage and timing of application 3. Demonstrate compatibility / synergy with existing programs 4. Finalize regulatory trials plans 5. Initiate trial program with partners and Key Opinion Leaders (KOL) on frozen TGAI and frozen formulation (Year 1)	North Hem.	30 to 50	100-200 m²	3,000-10,000 m²	500g - 1kg	
			South Hem.	30 to 50	100-200 m²	3,000-10,000 m²	500g - 1kg	Updated TEA with robust value proposition
Phase IV Pre-Launch Preparation	6	1. Perform Regulatory Trials for dossier submission (Year 1) 2. Continue trial program with partners and Key Opinion Leaders (KOL) on frozen TGAI and frozen formulation (Year 2) 3. Test new formulation options (for future products)	North Hem.	>50+	200-800 m²	1-4 ha +	1-10 kg +	Regulatory Field data for registration dossier
			South Hem.	>50+	200-800 m²	1-4 ha +	1-10 kg +	
	7	1. Perform Regulatory Trials for dossier submission (Year 2) 2. Perform Marketing / Pre-sales trials with KOL and commercial/distribution partners (Year 3) 3. Support regulatory dossier requirement (if needed) 4. Test new formulation options (for future products)	North Hem.	>50+	400-1,200 m²	2-6 ha +	10-100 kg +	
			South Hem.	>50+	400-1,200 m²	2-6 ha +	10-100 kg +	Regulatory Data Submission
	8	1. Perform Marketing / Pre-sales trials with KOL and commercial/distribution partners (Year 4) 2. Test new formulation options (for future products)	North Hem.	>50+	1,000-2,000 m²	5-10 ha +	100 kg +	
			South Hem.	>50+	1,000-2,000 m²	5-10 ha +	100 kg +	Potential approval in countries with rapid evaluation process
Phase V Launch & Market Expansion	9	Approval obtained : continue marketing / demonstration trials with multiple commercial partners and KOL in parallel to sales & distribution	North Hem.				> 100kg +	
		Approval obtained : continue marketing / demonstration trials with multiple commercial partners and KOL in parallel to sales & distribution	South Hem.				> 100kg +	Review and launch

Phases of Biomolecule Products Executive Overview

Phase 0
Product Concept

Phase 0 is the starting point where you will define the problem to be solved, the size of the opportunity, and current solutions to the problem (later this will be your positive control group). The key questions to ask for biomolecules are:

1. Is my technology adapted to the problem I am trying to solve?
2. Can I reach a valuable economic solution (based on a preliminary TEA?
3. How does my product differ from the existing best practices or solutions on the market in various geographies?

This phase should be heavy in interviews with farmers and customers of a future product.

Phase I
Pre-Field Discovery

Phase I or the "pre-field" phase of product development focuses on demonstrating the **efficacy of a novel lab or technology discovery**. This often includes screening biomolecule candidates and advancing leads through rounds of iterative design and testing (design-to-hit → hit-to-lead → lead-to-TGAI).

This is the proof-of-concept phase where the focus is on identifying the best lead candidates based on their efficacy, producibility and biological stability. This early stage serves to eliminate unviable options and establish a shortlist of candidate biomolecules worth advancing into further development. Exiting Phase I, a product should clearly demonstrate high efficacy under controlled conditions, with a good understanding of its MoA and be producible in a cost-competitive production system to demonstrate clear differentiation from existing solutions on the market.

Phase II
Early Product Development

Phase II focuses on **optimizing the lead biomolecule into its most efficacious product concept and testing it across multiple locations and conditions**.

Formulation development, use rate, and application timing are critical elements of this product optimization. In addition, production process development and scale-up studies should help prepare the selection of the best process steps and help refine and update the initial TEA to demonstrate viable COGS. Performing advanced studies to elucidate the MoA will increase the ability to implement a successful IP strategy. Finally, further assessment of any safety concerns will allow for preparation of the regulatory phase.

Phase III
Advanced Product Development

Phase III focuses on freezing the production process and the final formulation that will be selected for the regulatory dossier, as well as refining the **regulatory approval strategy**. Many studies during this phase will need to be executed at certified facilities that comply with testing standards recognized by regulatory authorities such as ISO standards, GLP3, and GMP 4. Part of the regulatory phase includes the registration of the production facility. During this phase, the production scale up at thousand-liter reactor volumes is usually realized with CDMO partners and represents one of the most critical steps to secure product quantity and quality for follow-on phases. Phase III also generally includes an increase in the reps and scale of field trials at a wider diversity of locations to confirm product efficacy data generated in earlier phases as well as establishing a product placement strategy. Engaging legal, manufacturing, and regulatory experts early and often is critical during this phase to ensure FTO and value capture for the product upon launch. In addition, early engagement with farmers/customers and affiliate organizations can lead to much easier Phase IV and Phase V experiences.

Phase IV
Pre-Launch Preparation

This phase will focus on **supporting the data packages and dossiers being evaluated** by regulatory authorities as well as preparation for the upcoming product launch. Simultaneously, a broad data set should be generated to support future marketing and sales efforts – product positioning. The focus should be not only on building up the data set but also on implementing a literature publication strategy, creating a product stewardship strategy, and developing an effective go-to-market plan. The supply chain strategy will also be developed for implementation in Phase V, selection of one or more large scale manufacturers as well as the regional location to minimize transportation costs.

Phase V
Launch and Market expansion

This launch phase focuses on **continual testing to support the business development** and expansion into new markets as well as the continual improvement of the product for improved efficacy and reduced COGS. This phase also includes continuing to work closely with legal and regulatory experts as the product is packaged, labeled, and sold, and looking for opportunities to extend the product lifecycle and maintain competitive position which can be done with novel formulations, product mixtures and application extension.

Comparison of Crop-Protection Biomolecules and Small Molecules

		Research			Development			
		Phase 0	Phase I	Phase II	Phase III	Phase IV	Phase V	Total
CP Small Molecule	Cost	$118-143m			$194-238m			**$312-381m**
	Time	3-5 years			7-8 years			**10-13 years**
CP Bio-molecules	Cost	$5-13m			$25-49m			**$30-62m**
	Time	1-6 months	2-5 years 6-18 months	18-36 months	24-36 months	5-9 years 24-48 months	12-24 months	**7-14 years**
Similarities		All the same	– Target selection and rationale-based design for de-novo design of Biomolecule compounds (RNAi, Antibodies, Protein Interaction, etc.) – In-vitro and in-planta screening approaches (efficacy test) – Sequence optimization	– Efficacy assays – Evaluation of spectrum of activity – Field trial scale up and testing	– Scaling up field trials to establish efficacy vs. Best-in-class positive controls and refining product positioning – Preparation for regulatory approval	– Conducting pre-market and regulatory field trials for submission – Engaging in proper product stewardship – Developing a go-to-market strategy with key partners	– Go-to-market strategies will be similar depending on the market – Customer and direct farmer engagement to drive belief/trust – Leveraging existing regulatory approvals to accelerate entrance into new markets	
Differences			– Bioprocess feasibility: "Process is the Product" – In-vitro to in-vivo translation: penetration can be a challenge – Different early-stage approaches for nature-derived compounds (ex. protein hydrolysate,	– Importance of biomanufacturing process development and transition from "active ingredient" to "TGAI" = Technical Grade Active Ingredient – The importance of encapsulation and formulation technologies for reaching the target of interest and ensuring good stability	– Production scale-up – specific to the bioprocess technology followed – Regulatory studies: Expected to be lighter, faster, and cheaper than with crop protection small molecules (based on current approvals). For biomolecules these studies either: – Benefit from a "special track" with a defined list of studies – Or follow a general track with the option to request multiple waivers to avoid the most costly and lengthy studies based on available scientific literature on this type of biomolecule – Differences in study preparation for analytical analysis due to the unique nature of biomolecules. Analytical chemists/biologists may need to adjust / develop appropriate analytical methods for this class of products	– Production scale up at commercial stage is different for biologicals than for chemicals in the sense that CMO are more specific and limited in this field and CAPEX investment for specific fermentations might differ and increase costs.	– Production considerations associated with shelf-life and transport – Labeling and safety requirements may differ depending on the region	

Glossary of Terms

Terms and definitions have been adapted from John Wiley & Sons Glossary of terms used in medicinal chemistry [21] for their applications in agriculture.

Active Ingredient	Molecule that provides direct biological activity or otherwise directly effects a target.
ADMET	Acronym referring to the absorption, distribution, metabolism, excretion, and toxicity profile or processes for a xenobiotic upon its administration in vivo. Note: ADME is also used to delineate these selected parameters within the context of a xenobiotic's chemokinetic profile.
AgTech	Short-hand term referring to Agricultural Technology; field of industry dedicated to the use of technology in agriculture with the goal of improving yield, efficiency, and profitability.
Allosteric	Site on a protein that can be bound by an effector molecule and is different from the protein's active site.
Assay	An investigative procedure or test to qualitatively and/or quantitatively measure the activity of research product.
Contract Manufacturing Organization (CMO)	Commercial organization that can be engaged to undertake specifically defined production of chemical or biological assets.
Contract Research Organization (CRO)	Commercial organization that can be engaged to undertake specifically defined studies.
Cost-of-goods sold (COGS)	All of the costs associated with manufacturing, delivery, and sale of a product.
Crop protection small molecule products	Made up of an active ingredient and other inert ingredients, formulated to be used as either seed treatments, in furrow, or as foliar applications to control pests and pathogens including weeds, insects, fungi, bacteria, and nematodes
Delivery	Process by which a crop protection formulation is administered to its intended target. Seed treatment, in furrow, and over-the-top foliar spray represent three delivery methods.
Experimental Design	How products are organized and tested in an experiment.
Field (In a Field Trial)	Made up of multiple plots in a usually contiguous piece of land.
Formulation	Mixture of active and inert ingredients that affects a target within an organism of interest. Herbicides, insecticides, fungicides, and nematicides represent a few formulations classes aimed at unique targets and organisms.
Freedom to operate (FTO)	In general, the ability to develop, make, and market products without legal liabilities to third parties. Relative to IP, FTO is the ability to develop, make, and market products without infringing the property rights of third parties.
Good laboratory practice (GLP)	Set of principles that provide a framework within which laboratory studies are planned, performed, monitored, recorded, reported, and archived. Note: These studies are undertaken to generate data by which the hazards and risks to users, consumers, and third parties, including the environment, can be assessed for researchers, agrochemicals, cosmetics, food additives, feed additives and contaminants, novel foods, biocides, detergents, etc. GLP helps assure regulatory authorities that the data submitted are a true reflection of the results obtained during the study and can therefore be relied upon when making risk/safety assessments.
Good manufacturing practice (GMP)	Quality assurance process that ensures that agrochemical products are consistently produced and controlled to the standards appropriate to their intended use.
Hit	Molecule that produces reproducible activity above a defined threshold in an assay.
Inert ingredient (inert)	Any ingredient intentionally added into a formulation that is not the active ingredient.
In silico	A process performed virtually.
In vitro	A process performed outside of a living organism (e.g. test tube, culture dish, etc.)

In vivo	A process performed or taking place inside a living organism
Intellectual Property (IP)	Intangible property rights covering inventions (patents), commercial indicators (trademarks), creative works (copyrights), and secret information (trade secrets).
Lead Optimization	The synthetic modification of a biologically active compound to improve the stereoelectronic, physicochemical, pharmacokinetic, and toxicologic characteristics of a hit for agrochemical usefulness and safety.
Location (In a Field Trial)	Larger geographic area usually with consistent weather pattern. Can be made up of many Fields.
Mode of Action (MoA)	Describes the mechanism for the activity resulting from the application of an active ingredient.
Pest	An organism or virus that can negatively affect crop growth or yield such as a weed, insect, nematode, bacteria, or fungi.
Plot (In a Field Trial)	Small unit of land on which a distinct test will be conducted. Plots can have many Rows.
Potency	The dose of active ingredient required to produce a specific effect of given intensity as compared to a standard reference. Potency is a comparative rather than an absolute expression of activity. A compound's potency depends on both affinity and efficacy.
Protein	Large biomolecules and macromolecules that comprise one or more long chains of amino acid residues and are encode by mRNA.
Row (many plants)	A defined line of plants grown in a single line of a plot.
Target	The component of a biological pathway thought to be of key relevance in an agricultural plant, pest, or disease, usually taking the form of protein, DNA, or RNA.

Organizations	Environmental Protection Agency (EPA) enforces the national policies around product safety associated with Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA)
	United States Department of Agriculture (USDA)
	U.S. Fish and Wildlife Service (FWS) enforces the national policies around product safety associated with the Endangered Species Act (ESA)
	Organization for Economic Co-operation and Development (OECD)
	National Marine Fisheries Service (NMFS) enforces the national policies around product safety associated with the Endangered Species Act (ESA)
	Food and Agriculture Organization of the United Nations (FAO)
	Food and Drug Agency (FDA) enforces the national policies around product safety associated with Federal Food, Drug, and Cosmetic Act (FFDCA)
	United Soybean Board (USB)
	National Corn Growers Association (NCGA)
	Western Growers Association (WGA)
	International Fresh Produce Association (IFPA)

Abbreviation	Definition
A	adenine
AI	Active ingredient
AMP	antimicrobial peptide
C	cytosine
CDMO	contract development manufacturing organization
CMO	[PLEASE DEFINE IN THE TEXT AND HERE]
COGS	cost of goods sold
CRO	contract research organization
de novo	Latin for ‘from the new’
DSP	downstream processing
dsRNA	double-stranded RNA
EC50	half-maximal effective concentration
EFSA	European Food Safety Authority
EPA	Environmental Protection Agency
EUP	experimental/emergency use permit [PLEASE CHECK]
FTO	freedom to operate
G	guanine
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
GRAS	Generally Recognized as Safe
HcAb	heavy-chain-only antibody
HPLC	high-performance liquid chromatography
IC50	half-maximal inhibitory concentration
In planta	Latin for ‘in the plant’
IP	intellectual property
ISO	International Organization for Standardization
LC	liquid concentrate
miRNA	microRNA
MoA	mode of action
mRNA	messenger RNA
PMO	project management office
RISC	RNA-induced silencing complex
RNAi	RNA interference
ROI	return on investment
ssRNA	single-stranded RNA
T	thymine
TEA	techno-economic analysis
TGAI	Technical Grade Active Ingredient
tRNA	transfer RNA
U	uracil
USP	upstream processing
WDG	water-dispersable granules

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