MIND THE GAP

BETWEEN UPSTREAM AND DOWNSTREAM BIOPROCESSING

Overcoming the challenges of bioprocess clarification and capture

MAGic™ BioProcessing

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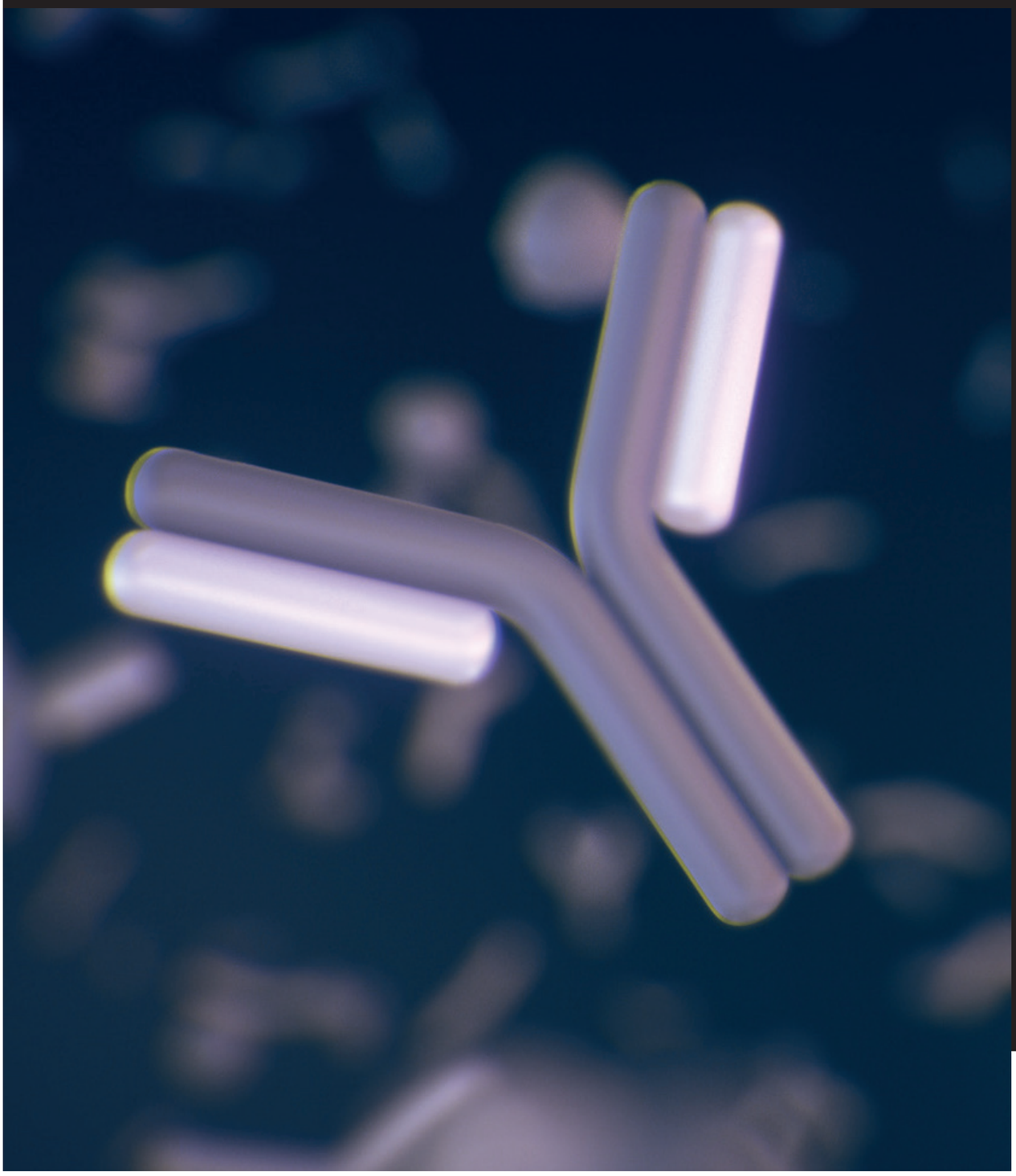


Mind the gap

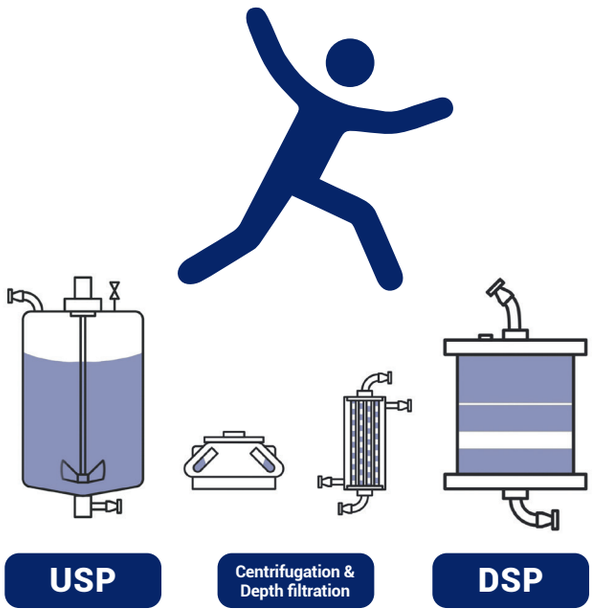
There is a gap between upstream and downstream bioprocessing. This gap reduces yield, throughput and productivity.

The gap is manifested and upheld by two legacy technologies: centrifugation and depth filtration. Neither upstream nor downstream, these orphaned technologies of cell clarification are rarely questioned by either side. Instead, they are often chosen out of habit and convention, rather than from a product or process quality perspective, despite creating bottlenecks in production.

Perhaps more importantly, the steps of cell clarification create a barrier between upstream and downstream processes because they are neither part of upstream's nor downstream's core objectives and goals. This barrier prevents integration of bioprocessing as a whole, which instead encourages separate optimization efforts. However, optimizing the parts does not necessarily create an optimized whole, sometimes it creates a Frankenstein's monster.

We argue that there is a need for a new cross-over technology to bridge this gap underscore the overall goal: better and safer end products.

This is what this ebook is about.



Note from the authors

This ebook has been created to highlight the need for innovation in bioprocess manufacturing, introducing a cross-over technology to integrate bioprocess for increased yield, throughput and productivity. However, we want to be clear about the fact that MAGic™ Bioprocessing develops and markets a scalable magnetic bead-based platform that allows for integrated clarification and capture directly from crude culture at any scale, thus replacing centrifugation, depth-filtration and protein A chromatography.

INTRODUCTION

Bridging the gap in bioprocessing

Over the past decades the biopharmaceutical industry has seen numerous significant advancements in productivity.

In upstream bioprocessing, high-density cell cultures from parallel large-scale bioreactors are producing increasingly higher yields and high-volume feed streams, while single-use technology and automation is increasing throughput and reducing turnaround times.

Downstream bioprocessing is continuously adapting to this ongoing upstream process intensification, involving both high-mass and high-volume feeds. As a consequence, downstream operations become increasingly complex, often necessitating additional measures such as dilution and concentration steps, as well as expertise in the intricate art of buffer management.

Despite the advancements in both upstream and downstream processing, the greatest challenges in biopharmaceutical manufacturing continue to be efficiency. Not only when it comes to novel modalities like cell therapies, but even for established therapies like monoclonal antibodies. Why is that?

The greatest challenges in biopharmaceutical manufacturing continue to be efficiency. Why is that?

It seems every solution to an existing problem presents a new problem. High cell density feed streams present new challenges for the clarification steps, often requiring dilution to reduce density or the addition of chemicals to promote flocculation and facilitate the clarification process. High volume product feed streams mean more eluent, more complicated buffer management, and larger buffer storage footprints. Low concentration of product means additional concentration steps, filtering problems and loss of throughput as massive volumes hit the chromatography racks, with resulting loss of yield and increase of HCP:s. And so on…

Clearly, there must still be untapped potential in bioprocess manufacturing, but are we even solving the right problems?

We suggests that the real problem in bioprocess is the gap between upstream and downstream. A gap manifested and upheld by the legacy technologies of the clarification steps i.e. centrifugation and depth filtration. However, these technologies are rarely questioned, because they are orphaned, operating on the borderline between upstream and downstream, tragically stuck between two different disciplines with two separate goals. As if they were children of estranged distanced parents with joint custody.

However, viewing upstream and downstream as two distinct processes often oversimplifies their primary objectives. Upstream's goal is framed as maximizing yields and product mass in crude culture, regardless of feed stream volume, while

A single unified technology that integrates the different needs of upstream and downstream is needed to bridge the gap in bioprocessing.

Downstream bioprocessing is a seemingly endless game of "follow the leader" as the upstream processes intensify.

downstream's goal becomes purifying that feed stream, no matter what comes their way and what challenges are encountered. A seemingly endless game of "follow the leader" as the upstream processes intensify, while downstream operations struggle to capture and purify, yet constrained by the same standard technologies and their inherent throughput flaws.

But consider this: what would it mean if upstream and downstream stayed true to the same overall goal, instead of two different subgoals? What would it mean for overall productivity, total costs and time-to-market, if the handover from upstream to downstream was smoother and more considerate? If the product was not only clarified but also less complex and significantly more concentrated when it hit the chromatography racks?

The only way to achieve this is to bridge the gap in bioprocessing byreplacing legacy clarification and capture technologies with a single unified technology that better integrates the different needs of upstream and downstream.

What goes upstream must come downstream.
The important question is: how much and when?

BIOPROCESS MANUFACTURING

Fix the bottlenecks in bioprocess to stay competitive

At first glance, the biopharmaceutical market, which has experienced rapid advancements in recent years, may seem to be maturing, with continued progress relying primarily on small, continuous improvements to the manufacturing process.

It is easy to believe that there is little room for true innovation in such a mature market, especially in some segments such as monoclonal antibody production. In fact, not so long ago, it was suggested that large-scale production facilities in combination with improved titers had resulted in a production capacity for mAbs that far exceeded the market demand over the coming years (Kelley, 2009). It was predicted that this would reinforce the use of already established production technologies, to reduce risk and that debottlenecking may have little practical value as the investment would never be recovered (Kelley, 2009).

We strongly disagree.

The notion of acceptably high efficiency is a self-fulfilling prophecy of failure

The market is growing fast and it is anything but mature. Continued strong market growth is fueled by the emergence of new therapies that address unmet medical needs, alongside a growing focus on preventative measures and specialized medications (Precedence Research, 2023). This not only requires the establishment of new production facilities and production lines to serve novel therapies and emerging regions, but also stresses the need to increase process efficiency. There are still several bottlenecks in bioprocessing. Many of the most critical ones originate from the gap between upstream and downstream processing. Thus, innovation and new thinking are not just opportunities but necessities for companies aiming to stay competitive.

The holy grail of biopharmaceuticals manufacturing is perhaps not the invention of a single new technology to save the day, but the ability of that technology to actually improve the complete bioprocess, bridging the gap between upstream and downstream to create a more unified, effective and reliable whole.

Any new technology and innovation that can unify the process has a natural place in the future of bioprocess manufacturing, because what goes upstream must come downstream. The important question is: how can we assure as much as possible does?

UPSTREAM BIOPROCESSING

Driving a race car in regular traffic

Upstream bioprocessing has seen numerous improvements over the past decades.

Each solution seems to present a new problem somewhere else in the process

Although, most of them with only one purpose in mind: to increase the amount of product mass in crude cell culture. Some of these improvements can be attributed to higher cell density and viability, but most of the increase comes from shear volume and throughput amplification from the use of multiple and larger bioreactors.

While this is completely in-line with upstream bioprocessing's core value process and goal, it doesn't always lead to increased overall productivity. On the contrary, given the challenges that the intensified feed streams present to downstream. Throughput problems, filter clogging, stoppage, maintenance, additional concentration and dilution steps, buffer management and storage problems are just a few examples of downstream problems that arise from upstream process intensification. Indeed, high titers from fed-batch cell cultures – with increasingly

Instead of applying ad hoc solutions for secondary problems we need to eliminate the true problem source.

higher cell densities and increasingly higher cell debris – has been described as a "curveball" that further complicates clarification the consecutive steps (Genetic Engineering & Biotechnology News, 2014). In fact, each solution seems to present a new problem somewhere else in the process. It begs the question: are we even dealing with the real problems?

Legacy technologies like centrifugation and depth filtration were incrementally improved rather than re-evaluated, despite presenting both direct challenges to clarification and secondary issues for downstream operations. Instead of pursuing an innovative cross-over technology, biopharmaceutical developers have been focusing on addressing the secondary problems arising from the increasingly complex downstream processes needed to manage high-volume and high-mass product feeds, such as additional pre-treatment steps, filter clogging, buffer management and an overall loss of throughput and yield. Meanwhile, the core problems of biopharmaceutical manufacturing still remain: productivity and throughput. Their cause and origin have only shifted from low titer problems to highly complex feeds and storage problems.

Instead of applying ad hoc solutions for secondary downstream problems, we need to eliminate the true problem source. A good start may be to question the legacy technology gap between upstream and downstream – centrifugation and depth filtration – and their consequences downstream.

Indeed, upstream's single-minded goal to improve the amount of product mass in crude cell culture no matter the output, may seem like a dangerous path as high-volume and high-mass feeds hit the chromatography racks. It's like driving a race car at full speed off the race track and into rush-hour traffic.

DOWNSTREAM BIOPROCESSING

What goes upstream must come downstream. The question is how much?

Upstream process intensification at the expense of downstream purification

Downstream capture and purification involve numerous steps, each adding to process complexity and reducing overall output. What goes upstream must come downstream. The question is how much, and that depends on the complexity of the process.

Although downstream principles have advanced over time, the underlying, well-established technologies suffer from limited throughput, and often require additional dilution and concentration steps when handling intensified upstream feeds. Clearly, downstream bioprocessing would become more efficient if upstream processes produced a less complex, more concentrated product.

However, rather than simplifying and focusing on the handover from upstream to downstream, i.e. the clarification steps, companies are adapting downstream operations to accommodate intensified upstream feeds, aiming to maintain productivity and throughput. Examples include buffer-on-demand systems and in-line buffer conditioning, which may reduce floor space and labor but offer limited solutions for process-related bottlenecks (DePalma, 2015, Lye, 2009), because when highly diluted feed streams hit downstream chromatography there will still be a need for additional pre-treatment steps and throughput problems.

All things considered, legacy clarification technologies create two types of bottlenecks in bioprocessing that need to be handled: facility-related and process-related bottlenecks. Facility-related bottlenecks, for instance, involve limitations in equipment size and process-related bottlenecks arise when consecutive steps in the workflow are mismatched in handling capabilities, leading to reduced process output (DePalma, 2015, Lye, 2009).

Replacing legacy clarification technologies and streamlining early downstream steps would provide opportunities for downstream and overall process intensification. A simpler downstream process would allow for fewer chromatography cycles, higher throughput, productivity and easier scale-up.

Clearly, the balance between upstream intensification, downstream capability and overall productivity is delicate. Not only does downstream processing require innovation to address the current challenges, but also some help from upstream and cell clarification. Although chromatography is commonplace and temperamental, it often represents the most expensive step in production (Schwaminger, 2019) enabling a clear economic benefit for alternatives in addition to robust and simplified workflows. Consequently, there is a need for complementing methods that improve overall productivity.

Legacy clarification creates both facility-related and process-related bottlenecks

CLARIFICATION

The bottleneck orphans of the gap

The bottleneck between upstream and downstream bioprocessing hinges on the centrifuge and the depth filter.

Centrifugation or depth filtration – a choice between a rock and a hard place

Consequently, the overall efficiency of modern bioprocessing depends on legacy technologies and their capacity to clarify increasingly complex feed streams, passing them on from upstream to downstream. However, a chain is only as strong as its weakest link, and in many cases clarification presents the most significant bottlenecks of bioprocessing. If these steps are inefficient, the entire process suffers.

The drawbacks of centrifugation and depth filtration

Clarification technologies present several direct drawbacks. In fact, both these technologies have been labelled "antiquated" and inadequate for modern bioprocessing (Genetic Engineering & Biotechnology News, 2014). Centrifugation, for instance, involves complex sample handling with a risk of contamination, is expensive, has limited scalability, and exposes sensitive biomolecules to high shear forces, leading to partial denaturation or aggregation, which can reduce yield and compromise product quality. Depth filtration, on the other hand, suffers from limited capacity, scalability challenges, and the risk of filter clogging and stoppage. Additionally, these clarification methods introduce

secondary problems downstream as they increase process complexity, adding steps that further reduce yield and throughput. Despite their drawbacks, these technologies have compromised a larger percent of the market.

The choice between these legacy technologies may seem like a choice between a rock and a hard place, but the situation is actually often worse than that. Since centrifugation at production scale only achieves around 90% clarification, a secondary depth filtration step is often needed (Genetic Engineering & Biotechnology News, 2014).

Bridging the gap

In this ebook, we have argued that the clarification steps disrupt and hinder overall process optimization, creating a gap that imposes significant bottlenecks, because they are not integral to the core objectives of either upstream or downstream. Bridging this gap is crucial, as it directly impacts overall productivity.

Upstream and downstream are mutually dependent yet often optimized independently, each operating on entirely different principles, with its own distinct goals and challenges, and each governed by different disciplines and managed by its own teams. This can lead to a lack of integration and alignment between the two stages, often resulting in inefficiencies and suboptimal process performance – because optimizing subprocesses does not by default optimize the overall process. Sometimes it creates a Frankenstein's monster.

Because upstream bioprocessing's biggest problem shouldn't be insufficient downstream bioprocessing – and downstreams biggest problem shouldn't be coping with increasingly complex, high-volume or high-mass feed streams that far exceed current technology capabilities. The balance between upstream intensification and downstream capabilities is delicate. Mismatches between the two lead to bottlenecks, overloading downstream purification capacity, or underutilization of resources. As the process becomes increasingly over-complicated with more steps, the overall yield drops.

The paradigm of the centrifuge and depth filtration has created a glass ceiling that limits the productivity of contemporary bioprocessing

Unleash productivity in bioprocessing

It's time to reexamine the bottleneck orphan technologies of clarification and move towards a more holistic bioprocessing approach that optimizes overall yield and productivity. Replacing centrifugation and depth filtration with a disruptive crossover technology is essential, as the current clarification paradigm has imposed a ceiling for productivity in modern bioprocessing.

To achieve this, we must abandon the misconception that biopharmaceutical markets, such as monoclonal antibody production, are mature and saturated, with little room for truly new innovation to yield returns. Instead of focusing on minimal-risk process variations and choosing operational setups based on factors like footprint, space constraints, existing agreements, and even habit, we need to challenge the legacy technologies of clarification and actively pursue new disruptive crossover technologies.

A better way

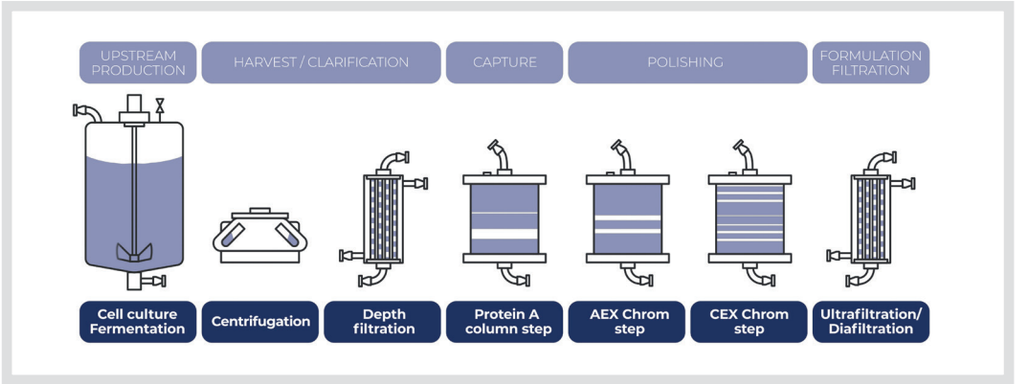
One potential solution is magnetic separation that can replace both centrifugation, and depth filtration to enable one-step clarification integrated capture and recovery of product, directly from crude cell broth. Thus, eliminating bottlenecks and simplifying downstream processing to improve overall productivity.

Skip a step. Gain a leap.

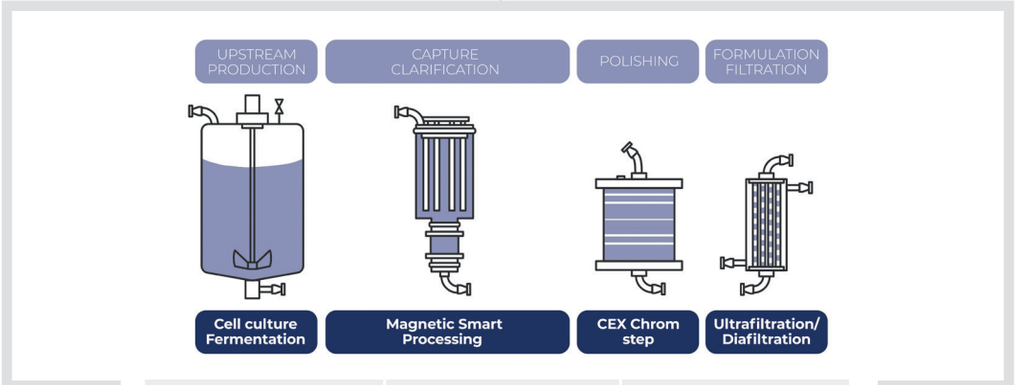
MAG-SP is a magnetic bead-based platform for 1-step clarification and capture of biological modalities directly from crude cell broth. As the world's first scalable high performance magnetic separation solution, thanks to the best-in-class paramagnetic characteristics and large surface area of the robust resins, it truly sets a new standard in bioprocessing – at any scale, from research and pilot to full manufacturing.

MAG SP is a cross-over technology that better balances the different goals and challenges of upstream and downstream for significantly increased yield, throughput and productivity at lower cost. By replacing conventional centrifugation, depth filtration and capture with a single step, the platform reduces process complexity and eliminates bottlenecks.

Column based process



Magnetic smart process

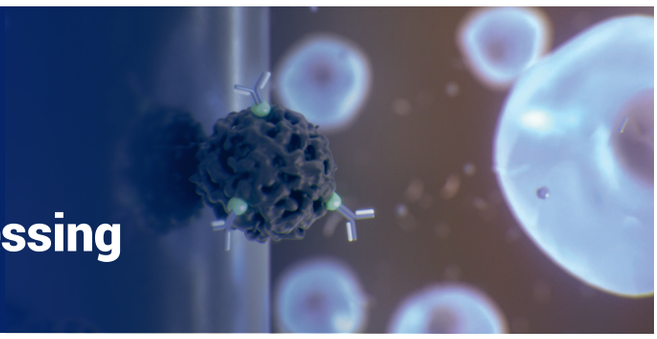


+ 34 % Yield	+ 40 % Throughput	48 %* Productivity
- 38 % COGS	- 57 % Water usage	< 10 ppm HCPs

*Brechman et al. (2024) 'Internal study, MAGic Bioprocessing'.

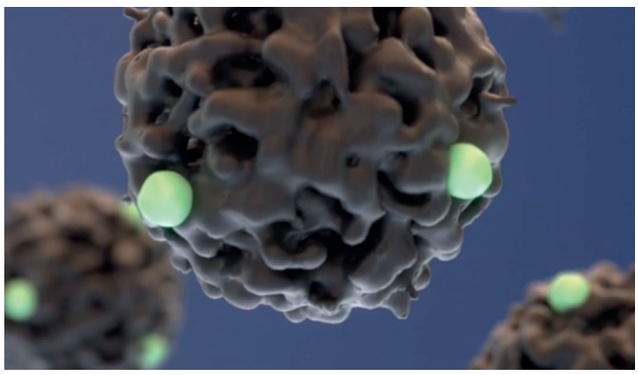
MAG-SP

MAGnetic Smart Processing



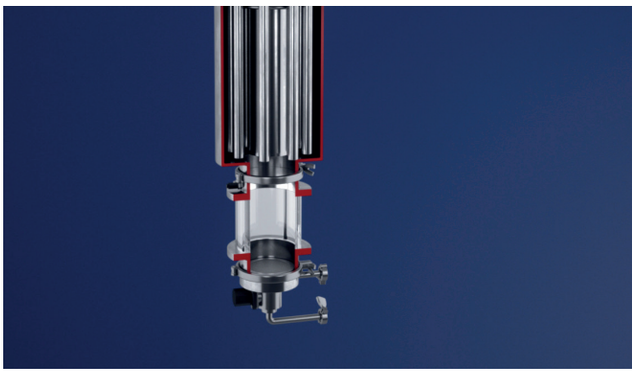
MAGic™ Beads

At the heart of the platform lies our proprietary superparamagnetic bead technology based on high-surface agarose resins, customizable for a variety of applications, including monoclonal antibody capture.



MAGic™ Accio systems

Paired with our fully automated, state-of-the-art magnetic separator systems, the platform ensures rapid, gentle and effective magnetic separation with a small footprint.



MAGic™ Kits

Evaluation kits for testing our platform on your application area.

Discover how **MAGic™ Accio** works. Scan the QR code to see the video!

SUMMARY

A new paradigm in bioprocessing

Unquestioned legacy clarification technologies like centrifugation and depth filtration are hampering productivity and causing downstream problems to capture and the consecutive chromatography steps.

These orphan technologies are causing a gap between upstream and downstream bioprocessing that prevents optimization of the process as a whole, because they are neither part of upstreams nor downstreams core process and goals. The anomalies are mounting up, delivering a sad story of inefficiencies and unnecessary bottlenecks.

Magnetic separation offers a gentler and more effective way of achieving cell clarification and protein capture in a single step, directly from crude cell culture, replacing both centrifugation, depth filtration and Protein A column chromatography.

Unquestioned legacy clarification technologies like centrifugation and depth filtration are hampering productivity and causing downstream problems to capture and the consecutive chromatography steps.

Isn't it time for a new paradigm in bioprocessing?

– one that addresses the root causes instead of relying on ad hoc solutions to mask the symptoms.

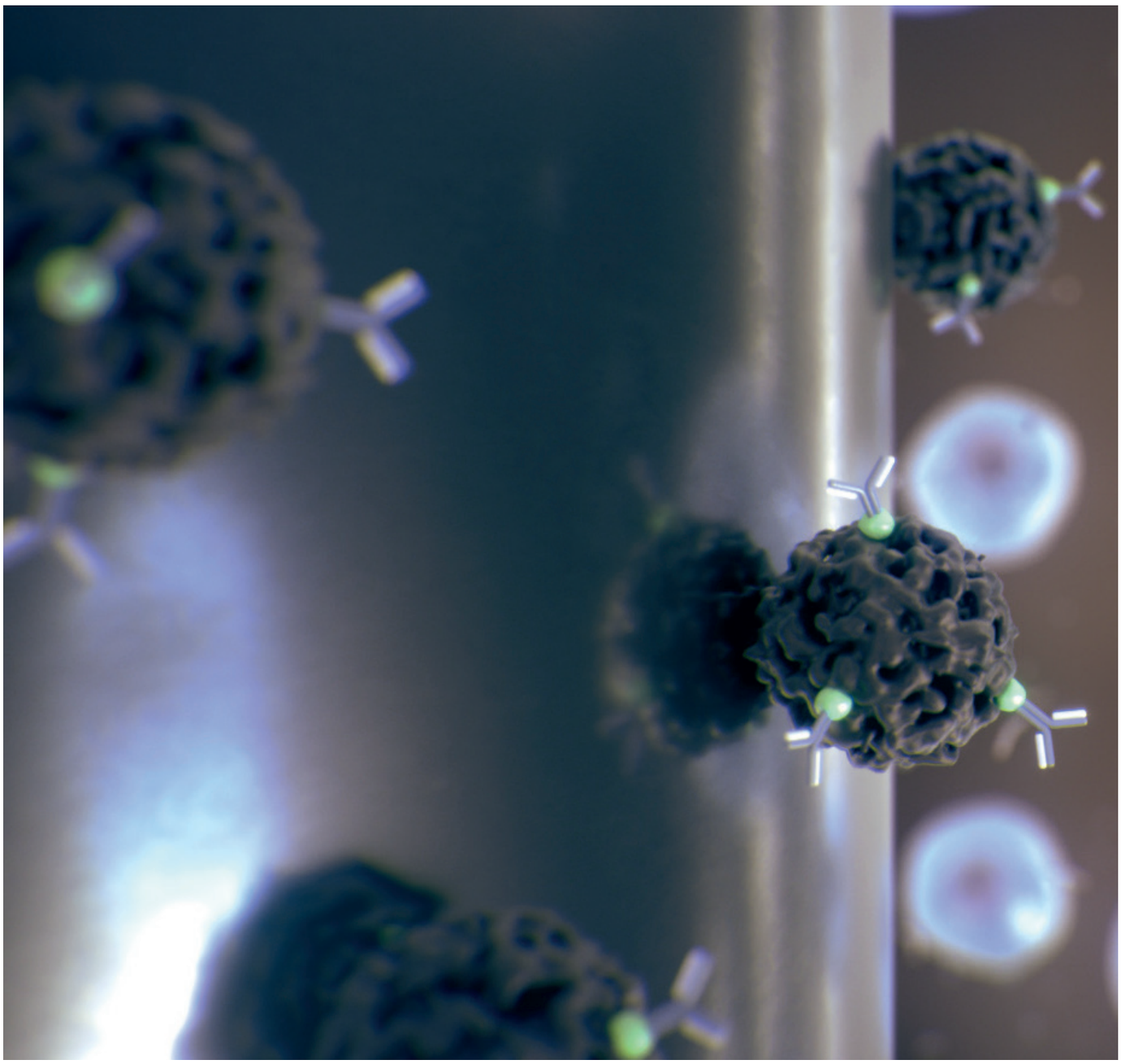
MAG-SP is the world's first scalable magnetic separation platform, utilizing super-paramagnetic, high surface, agarose beads. Studies report up to 34% yield increase, 40% boost in throughout increase and a 38% reduction in COGs with low HCP and drastically reducing buffer and water usage.

Skip a step to gain a leap in throughput, productivity and yield by introducing a disruptive cross-over technology that bridges the gap between upstream and downstream to align their goals and eliminate key bottlenecks.

Do you want to know more?

Do you want to know more about our **MAG-SP platform**,our magnetic seperator, **MAGic™ Accio,** and our magnetic beads, **MAGic™ Beads** mAb, for more efficient monoclonal antibody production?

Visit www.magicbioprocessing.com

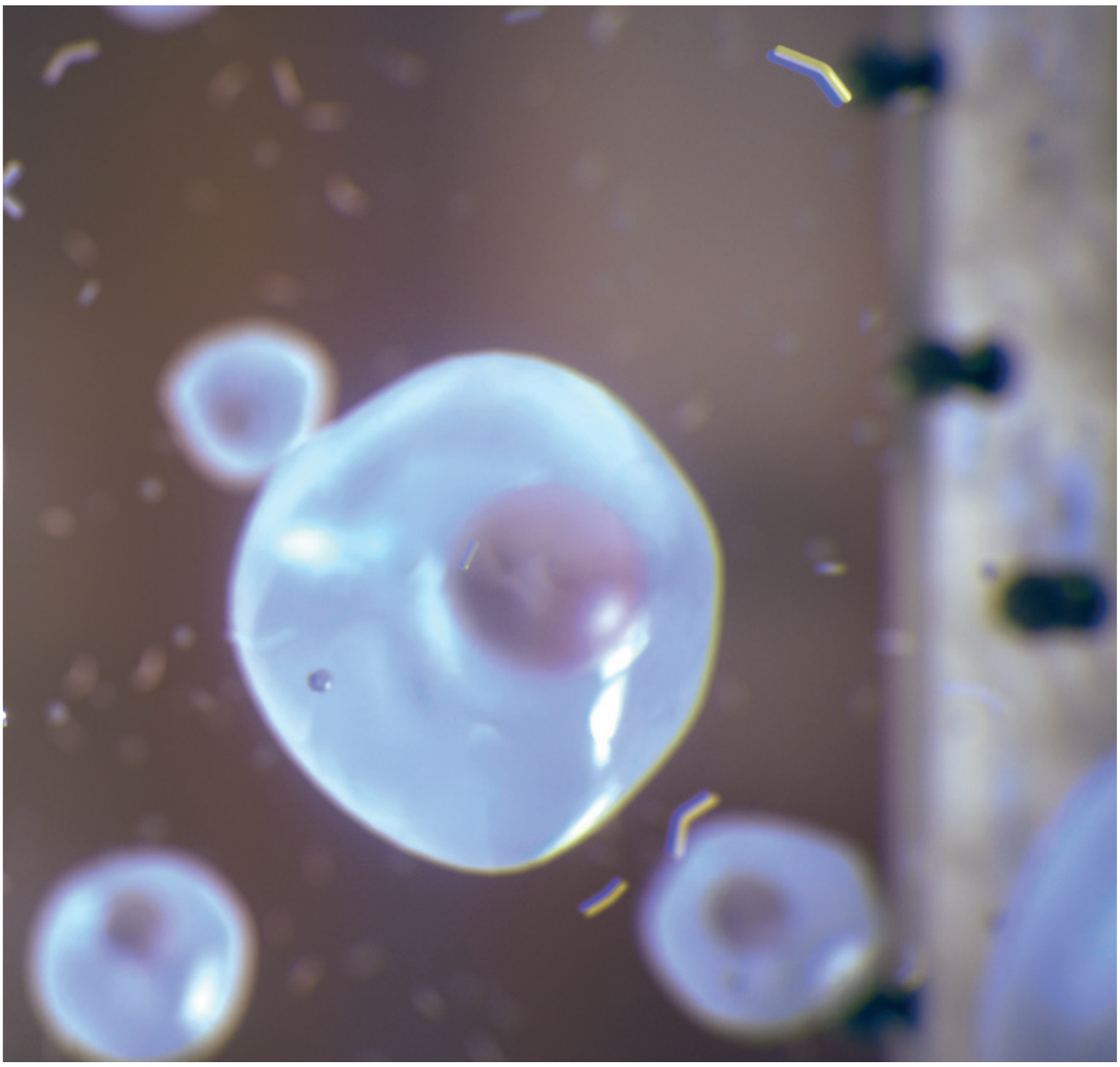


MAGnetic Smart Processing **(MAG-SP)** is the world's first truly scalable magnetic separation solution, enabling 1-step clarification and capture of biological modalities directly from crude cell broth.

Let's talk!

Contact us to discuss how our products fit your need, or even find solutions to your own custom applications.

Mail: info@magicbioprocessing.com



Learn more!

Would you like to know more about our **MAG-SP platform**, our magnetic seperator, **MAGic™ Accio,** and our magnetic beads, **MAGic™ Beads** mAb, for more efficient monoclonal antibody production? Do you have your own custom solution you would like to investigate with us?

Visit www.magicbioprocessing.com

Discover how **MAGic™ Accio** works. Scan the QR code to see the video!

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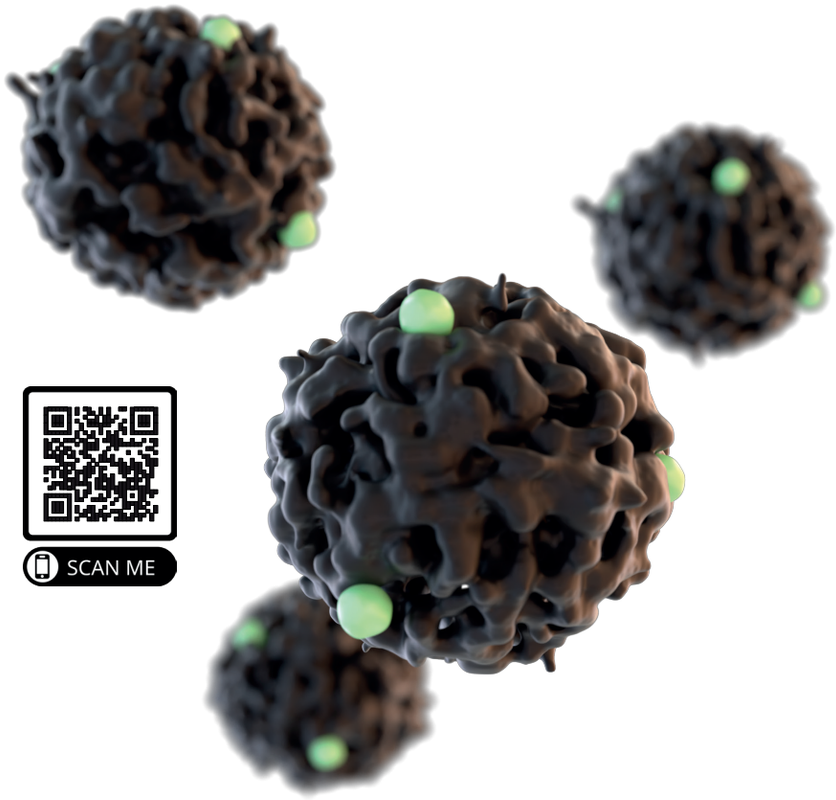

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