

**Sterling** 



# Biocatalysis:

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Leveraging nature's catalysts in  
pharmaceutical development



# Introduction

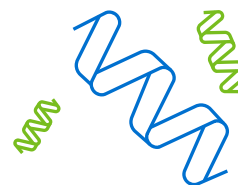
Enzymes are considered nature's catalysts. They are found in all living organisms and aid in specific natural processes such as digestion and DNA replication.<sup>1</sup> Over the years, various industries have found ways to apply enzymes to synthetic chemistry, finding ways to harness their catalytic activity in a controlled environment. In fact, although people have used enzymes in brewing for thousands of years, they didn't know what enzymes were or understand that they could be isolated until the 1800s.<sup>2</sup>

As the scientific field advanced, the application of enzymes expanded far beyond brewing, with scientists only beginning to realise the full potential of enzymes in the late 20th century. As they developed the ability to evaluate an enzyme's suitability for various processes, Biocatalysis, the use of enzymes to speed up chemical reactions, began to gain traction. Today, Biocatalysis is employed within a diverse set of industries, including food and beverage, nutraceuticals, pharmaceuticals, biotechnology, and more. In 2019, it was estimated to have several hundred commercial applications.<sup>3</sup> For pharmaceutical development, Biocatalysis can be particularly compelling as it offers superior enantio-, regio- and chemoselectivity to chemical catalysis, whilst maintaining other benefits such as efficiency and cost-effectiveness. Furthermore, enzymes mostly work under very mild conditions, though there are exceptions that work under harsh conditions.

**In this paper, we will closely examine the growth of Biocatalysis in pharmaceutical development, the power of enzyme immobilisation to overcome some of Biocatalysis' key challenges, and why Sterling's partnered approach to biocatalysts provides solutions for our customers.**

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The global market for biocatalysts and Biocatalysis is expected to **grow 15% between 2019 and 2024.**<sup>4</sup>

# The growth of Biocatalysis

## **If enzymes are naturally occurring and have been used for centuries, why is the widespread use of Biocatalysis in manufacturing relatively new?**

The primary reason is that advances in technology have enabled far more widespread access to natural enzymes, as well as the controlled optimisation of natural enzymes for greater catalytic efficiency. This, coupled with more widespread awareness of Biocatalysis' compelling advantages, are collectively responsible for the technology's rapid growth.

**Let's take a closer look.**

Over the past 20 years, next generation DNA sequencing has revolutionised genomic research. Ultra-high throughput capabilities have allowed genetic material to be read faster than ever before. This data has been harnessed in open-access databases such as TrEMBL, allowing researchers to rapidly progress the area of Biocatalysis.<sup>5</sup>

Furthermore, inventive methods of DNA manipulation have allowed enzymes to be customised in a highly controlled manner. These methods include directed evolution and protein engineering, which now make it possible for enzymes to be tailored and optimised for a given chemical reaction. In 2018, American chemical engineer Frances Arnold was awarded the Nobel Prize in chemistry for her work in directed enzyme evolution.<sup>6</sup>

Finally, the expanding field of enzyme immobilisation has yielded highly efficient, robust and stable heterogeneous biocatalysts – tackling some of the key challenges related with the use of enzymes at scale.

## **Coupled with enzyme commercialisation, four compelling advantages have driven the growth of Biocatalysis:**

### **Efficiency:**



In one example, the enzyme carbonic anhydrase was shown to catalyse the hydration of 4 million molecules of CO<sub>2</sub> per second, approaching the limit of turnover dictated by diffusion.<sup>7</sup>

### **Cost-effectiveness:**



Biocatalysis avoids the need for scarce precious metals and eliminates the cost of removing them.<sup>8</sup>

### **Sustainability:**



Biocatalysis aligns with key green chemistry principles, like using renewable resources and generating residues that are not environmentally harmful.<sup>9</sup>

### **Customisation:**



In one example, scientists were able to produce the HIV drug islatravir using solely enzymes, 5 of which were engineered.<sup>10</sup>

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**Nearly 1000 (983) new classes of enzymes have been discovered since 2017.** <sup>11, 12</sup>

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# Immobilisation for enzyme optimisation and risk mitigation

In spite of its promising benefits, the pharmaceutical industry has been relatively slow to adopt Biocatalysis in drug development until recently. Three main concerns with utilising enzymes have included their high upfront costs, challenges detecting and separating them from products, and their sensitivity to changes in the reaction environment. **The question, then, is what can be done to alleviate these key concerns when incorporating Biocatalysis into pharmaceutical development?**

Enzyme immobilisation, or the practice of affixing to or encapsulating an enzyme within a heterogeneous solid support, presents a promising solution to all these major challenges. There are a number of methods for immobilisation, including adsorption, ionic bonding, covalent bonding etc.; and immobilisation supports or “media” are most often polymers or inorganic materials, such as polyacrylates or silica.<sup>13,14</sup>

When attached to a solid, the **stability** of an enzyme to conditions such as temperature, pH and organic solvent content can be significantly improved. In one example, an immobilised  $\alpha$ -chymotrypsin was shown to be 1000 times more active in a methanolic environment compared to its free enzyme counterpart.



PROBLEM	SOLUTION
<b>Changes in the reaction environment</b> can result in loss of enzyme activity.	Immobilisation creates a stable microenvironment for enzymes to operate in, <b>making them stable to changes in reaction conditions.</b>
<b>Enzymes can be challenging to remove</b> from end products.	By being attached to a solid, <b>enzymes can be easily separated</b> from the final product via simple filtration or centrifugation.
Even with modern manufacturing techniques, novel <b>enzymes can be expensive to source.</b> <sup>15</sup>	Immobilised enzymes can be easily separated, stored and recycled. <b>Reusing immobilised enzymes offsets acquisition costs.</b>

Secondly, immobilisation can help to ensure **product purity**. Attachment to a solid makes the enzyme easier to separate from the desired drug after the reaction is complete. **Commonly used methods of enzyme recovery include simple filtration or centrifugation.** Pharmaceutical developers can take comfort in knowing that their end products have been purified through these reliable and cost-effective methods.

Finally, because free enzymes are challenging to separate and isolate from completed reaction mixtures without enzyme denaturation, they are often only used once before disposal.<sup>15</sup> This is inefficient and wasteful, diminishing the cost-effectiveness of Biocatalysis for the pharmaceutical sector and other industries.

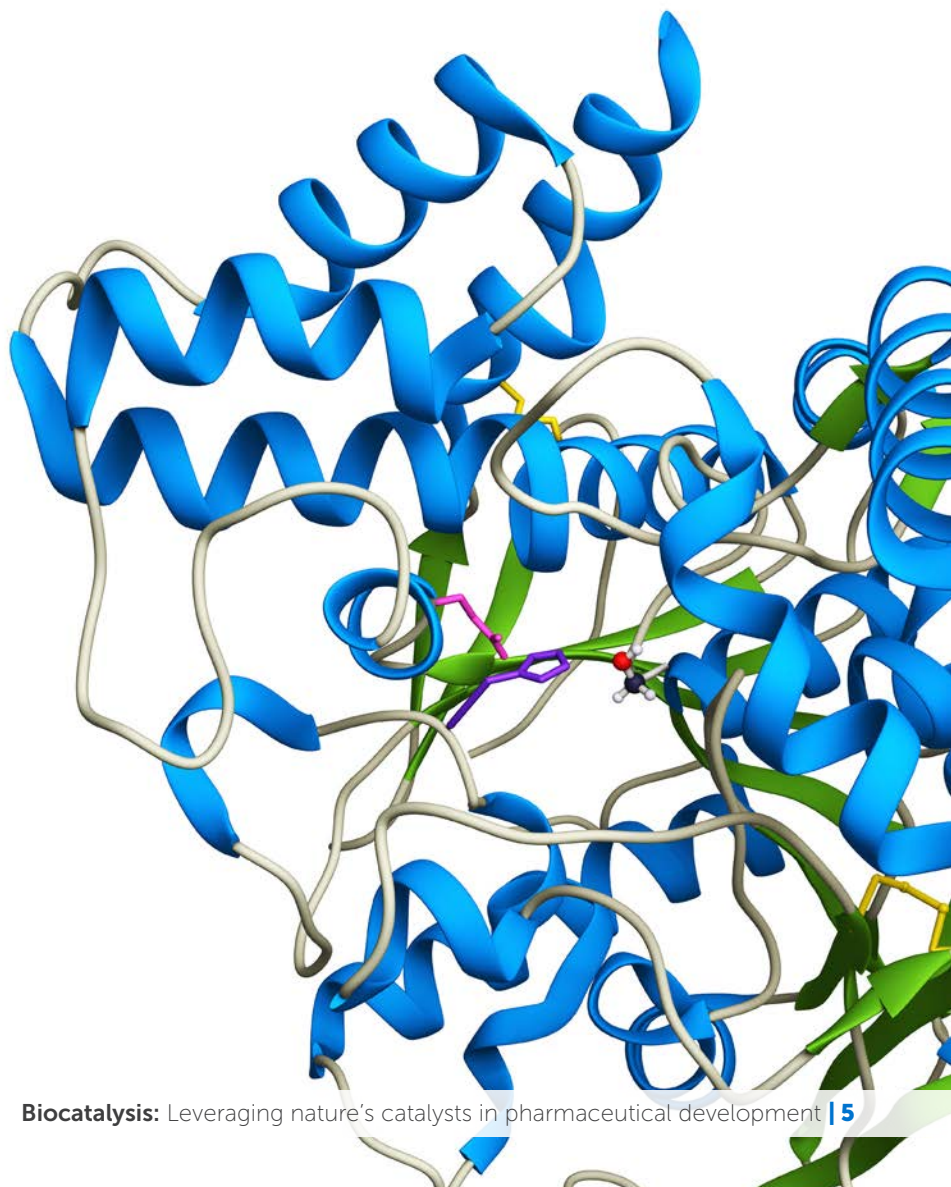
As immobilised enzymes can be readily separated and stored, however, they may be recycled and reused over many batches to **reduce overall costs**. **Immobilised lipases, for example, have been recycled over 20 times on a current customer project here at Sterling, without significant dimishment of enzyme activity.**



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In a customer project at Sterling, **a lipase was immobilised onto 15 different immobilisation media, with significant retention of enzyme activity.**

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# Sterling's partnered approach to Biocatalysis

**At Sterling, we have decades of experience working with complex chemistries, including the use of enzymes.**

We are committed to continually embracing new technologies that have the power to bring significant advantages to the API development and manufacturing process.

We have over 50 years of experience taking novel chemistry from the laboratory to industrial scale. We grant you full ownership over the IP that results from the enzyme screening and development process, and we couple our enzyme expertise with full cGMP manufacture of APIs and intermediates.

Lastly, we are a partnership development and manufacturing organisation (PDMO). Our differentiated commitment to transparency and simple, collaborative ways of working is instrumental to providing our customers peace of mind as they embrace Biocatalysis.

Our Technology and Innovation Programme underscores our commitment to continual innovation and improvement. To complement and expand our own capabilities, we have carefully selected experienced partners to maximise value throughout the entire Biocatalysis process, from suitability screening to full scale-up.

**Our partners represent some of the world's leading academic and commercial expertise in Biocatalysis. They include:**



**Northumbria University**  
NEWCASTLE

Northumbria University, particularly their Nzomics business unit, assists with rapid enzyme screening, customisation and optimisation through directed evolution, and immobilisation.



**UNIVERSITY OF LEEDS**

The Centre for Doctoral Training in Molecules to Product helps us provide our customers with the significant manufacturing advantages afforded by immobilisation.

**Sterling**

**Would you like to learn more about our world-class Biocatalysis solutions?**

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