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Precision Fermentation: A Molecular Mimic for Animal Protein

(Review Article)

Abstract

The conventional animal agriculture industry faces significant challenges including environmental degradation, ethical, and social concerns, and the growing global demand for proteins. Precision fermentation is a cutting-edge technology, that offers a promising approach to producing animal proteins without the need for traditional animal husbandry. This review interprets the principle of precision fermentation, utilizing genetically modified microorganisms to produce specific proteins by introducing target genes, and illustrates the current applications of precision fermentation in animal protein production. A range of meat proteins, including collagen, hemoglobin, myoglobin, dairy proteins, such as lactalbumin, lactoglobulins, caseins, lactoferrin, whey protein, and egg white proteins have been produced with the use of the technology, which the review demonstrates their applications in various animal product processing companies worldwide, through their products in meat substitutes such as plant-based burgers, egg white proteins in processed foods; ice-cream, bakery items, milk, cheese and dairy alternatives such as cheese, ice-cream, yogurt and frozen dairy products, functional ingredients, flavoring agents and fat types in replacing popularly demanded animal-based products. The review overviews their production, market availability, functional properties, and end uses. Furthermore, the review highlights the advantages and challenges associated and the regulatory aspect of the technology in replacing animal proteins.

1. Introduction

By 2050, the world's population is expected to reach about 9.7 billion, about one-third increase from 2015. As cited in, Henchion et al., (2021), to meet the demands of the expanding population, the amount of protein that must be produced in 2050 must be 80% greater than it is now, assuming current intake levels. About 65–80% of the protein consumed in Western countries, now comes from dairy and animal products (Auclair & Burgos, 2021).
Livestock production is a significant cause of environmental issues about biodiversity, freshwater supply and pollution, land degradation, and climate change (Joyce et al., 2012). Around 95 million animals in the United States are fed by the more than 835 million acres, or 40% of the total land area, that are utilized to cultivate livestock feed. Of these, 630 million acres are used for beef and dairy cattle alone (Tubb & Seba, 2021). Due to its substantial impact on climate change, enteric methane (CH_4) emissions from farm animals are receiving international attention. Increasing the number of animals and land used to produce more protein is not sustainable (Nielsen et al., 2024). According to Garnett, (2009), meat and dairy products are the foods that have the biggest environmental impact, contributing to roughly half of food-generated greenhouse gas emissions and, in fact, 18% of worldwide GHG emissions.

In addition, there has been a noticeable change in consumer preferences in recent years toward healthier options, as well as an increase in knowledge of the ethical concerns and animal welfare related to animal products (Knychala et al., 2024).

Achieving a delicate balance between reducing the environmental impact of present farming techniques and expanding food production to fulfill the demands of a growing population is necessary to address these issues. Consumer views have changed, and environmental concerns about animal products have sparked interest in and investment in the innovative field of cellular agriculture (Knychala et al., 2024). An innovative method that mimics animal proteins without the environmental impact of livestock farming is precision fermentation. As cited in Chai et al., (2022), fermentation is the chemical conversion of any organic material by microbial metabolism, which is facilitated by a variety of enzymes.

Genetically modified production is the technology underlying precision fermentation. The microorganism, which is typically grown in a bioreactor, needs sugar, nutrients, and air to grow and create the desired protein. The product is collected, partially purified, and prepared for direct use as a food ingredient at the end of fermentation. The microorganism, the fermentation process, and the protein's physicochemical characteristics all influence the methods used for protein recovery and purification (Nielsen et al., 2024). *Yarrowia lipolytica, Aspergillus oryzae*, *Kluyveromyces lactis, A. niger*, *K. phaffii* (previously *Pichia pastoris*), *Saccharomyces cerevisiae,*

T. reesei, and a few bacterial species (*Bacillus*) are the most frequently used species for precision fermentation of food-related products at industrial scale (Chai et al., 2022).

A number of issues with the current global manufacturing system may be resolved by precision fermentation. Precision fermentation has been demonstrated to have a better environmental impact than animal products; certain crops require about 90% less land and 96% less water (Hassoun et al., 2024). According to life cycle assessment studies, precision fermentation can produce GHG emissions that are 97% lower than many standard production methods, particularly those that use animal sources (Järviö et al., 2021). Furthermore, precision fermentation removes the need for animal slaughter, resolving ethical issues surrounding animal welfare and lowering the possibility of antibiotic resistance and zoonotic illnesses, two serious public health problems related to intensive livestock farming (Elsohaby & Villa, 2023).

The capacity to scale up to manufacturing production is a crucial component of precision fermentation's success. Significant investments have lately been made in the precision fermentation business. 382 million USD of the 842 million USD raised by fermentation firms in 2022 went toward capital investments in precision fermentation (Nielsen et al., 2024). Since the 1980s, human insulin, growth hormones, enzymes like rennet (chymosin), and other biological products have been produced commercially using precision fermentation, a proven method. This method is used almost entirely to create a large number of vitamins and supplements (Knychala et al., 2024).

The potential of precision fermentation to transform food production is examined in this review, along with its uses, advantages, challenges, and potential for reducing dependency on animal- based diets.

2. Impacts of Animal-based Agriculture

2.1. Environmental Impacts

Significant attention has recently been directed toward the environmental impacts of animal-based agriculture, which is believed to be responsible for 26% of global greenhouse gas emissions; over half of these emissions, notably, stem from livestock farming alone. Therefore, research on animal agriculture is essential to meeting global climate goals. An estimated 11–20% of all greenhouse gas emissions are attributed to the cattle industry—consequences that extend beyond just carbon emissions, encompassing waste pollution, deforestation, and biodiversity loss. Meat, dairy, eggs, and aquaculture products account for around 83% of the world's agricultural land. These outputs, however, only provide 37% of the world's protein supply and 18% of its calories, indicating a serious mismatch between resource input and food yield. A major cause of biodiversity loss, this excessive resource usage is linked to over 80% of global deforestation and contributes to the degradation of natural habitats and the extinction of species (Clark et al., 2017). The waste generated in livestock and poultry operations presents considerable risks to water quality and soil health, in addition to the impacts of land use and greenhouse gas emissions. If these activities are not managed appropriately, they may harm water sources by producing manure, bedding, milk house wash water, spilled feed, and mortalities. The presence of nutrients such as nitrogen and phosphorus in manure can lead to eutrophication and an increase in biochemical oxygen demand in aquatic systems because they leak into groundwater or run off into surface waters, particularly in regions with limited land area for nutrient absorption. The location of livestock facilities influences the potential for pollutant runoff; in grain-deficient areas, nutrient accumulation often exceeds acceptable limits. Another concern with restricted animal operations is air quality. Recent research is focusing on the health and environmental consequences of particle emissions, including dust, ammonia, and hydrogen sulfide, which can adversely affect respiratory health and contribute to environmental degradation. Although it has historically focused on odor problems, however, the broader implications are becoming more apparent. A comprehensive strategy for environmental management and resource efficiency in animal-based agriculture is needed to address these effects; however, this necessity is often overlooked (Abdalla et al., 2006). Because of the significant impact on ecosystems, it is crucial to develop effective practices. Although there are various approaches available, not all are equally viable. Thus, attention must be paid to the sustainability of these methods, but the complexity of the issue complicates the implementation.

2.2. Social Impacts

Animal-based agriculture has massive social impacts, such as those on human health, religious sensitivities, and the surge in veganism/vegetarianism that we observe today. The risks to our health posed by animal husbandry, such as the development of resistant bacteria due to the non- therapeutic use of antibiotics in livestock, have been well known for some time and represent a global public health problem. Particulates and harmful gasses like hydrogen sulfide and ammonia are also released from concentrated animal feeding operations (CAFOs), which are associated with cardiovascular and respiratory problems for both employees and nearby communities. In the discussion of animal agriculture, human health is a critical topic of concern because these emissions are linked to respiratory conditions, an increased risk of developing asthma, and an increased exposure to zoonotic infections (Abdalla et al., 2006).

Animal agriculture also has a root in religious concepts. Religious beliefs affect the way animals are slaughtered and treated it can be noted in halal and kosher traditions or the limit of meat from a certain species of animal, like cows or pigs for some groups. Under these circumstances, livestock farming is forced to obey the religious dietary laws Kosher and Halal, therefore affecting both production manners as well as market dynamics. While these rules have changed the way animals are allowed to be processed, they also raise ethical and welfare questions about how we treat living things, leading some people not to eat animal products at all (Sullivan et al., 2013). In recent years, additionally, in parallel, there has never been as much interest and demand for veganism/vegetarian ways to eat in society due to ethical reasons, health reasons, or environmental ones. Consumers are falling out of love with animal agriculture, and for good reason—the greenhouse gas emissions, inefficiencies in resource uses like water, and the offensive way treat animals. There are appeals from animal cruelty and the sustainability movements that have seen a worldwide shift of people turning towards plant-based eating. This trend has implications for animal agriculture by adding market pressure to find plant-based substitutes and is indicative of a wider interest in sustainability and transparency in food production (Saitone et al., 2006).

2.3. Ethical Impacts

In particular, animal welfare has drawn attention to the moral dilemmas raised by reliance on animals as workhorses or food suppliers. Furthermore, viewpoints that stress ethical treatment of animals and animal rights urge that modern intensive farming methods, which often put productivity before animal welfare, be subject to radical critique. Animals reared in such concentrated environments as confined animal feeding operations (CAFOs) are subject to physical and mental stress, and their natural activities are restricted. Animals of the cloister shed entirely lack an environment appropriate for them, a situation that, although not inherently cruel, constantly makes the animals unhappy—mediocre at best (Pond, 2011).

The impact of these practices on the quality of life for animals has sparked concerns about their morality. Common procedures, such as tire cutting and castration, are sometimes performed without anesthesia to minimize risk to humans or because the tools are easier to manage at this stage—though this practice is also debated as unethical. While many of these methods are standard in animal husbandry, they highlight the urgent need for improved welfare legislation and raise worries about unnecessary suffering. In light of these criticisms, the field of animal welfare science has emerged as a new discipline focused on developing more humane techniques that balance ethical treatment with production efficiency. Some stakeholders believe that providing enriched conditions where animals can exhibit their natural behaviors and receive proper veterinary care is crucial to ethical farming, and this development has led to proposals for rules and regulations that mandate these requirements (Dawson et al.,2005).

The economic factors affecting industrial agriculture add another layer of complexity to animal welfare concerns. The focus on efficiency often takes precedence over the well-being of animals, driven by the strong demand for animal products. This can lead to cost-cutting practices that may compromise the care animals receive. Ethical considerations in animal agriculture emphasize that the moral obligation to treat animals with respect and reduce their suffering should not be overshadowed by financial pressures. As a result, customers are supporting animal welfare certifications that highlight humane production methods at all stages and are calling for products obtained responsibly (Sullivan et al., 2013). Furthermore, a wider philosophical discussion regarding the relationship between humans and animals has been sparked by the moral dilemmas surrounding animal care in agriculture. Certain ethical theories, such as utilitarianism and rights- based approaches, suggest that animals should be acknowledged as sentient beings with inherent value rather than simply viewed as resources. This viewpoint has played a significant role in the growing popularity of vegetarianism and veganism, as more people aim to lessen the suffering that animals endure in farming practices (Pond, 2011). As a result, the moral dilemmas associated with animal-based agriculture underscore the pressing need for updated industry standards and clear regulations that focus on animal welfare. Enhancements in these areas could help address some ethical issues and foster a more compassionate approach to food production that resonates with changing consumer values.

3. Precision Fermentation Technology

3.1. Concept

Growing awareness of the ethical issues surrounding animal products (Frank, et al., 2022), the shift in consumer preferences toward healthier options, the effects of climate change, and population growth predicted to surpass 9.7 billion people by 2050 are all contributing factors to the challenges facing the global food production system (Knychala, 2024). A fine balance must be established in order to address these issues: raising food production to satisfy the demands of an expanding population imposed by current practices of agriculture. At the same time, changes in consumer perceptions and concerns about animal products' effects on the environment have led to interest in and investment in cellular agriculture, an innovative field. The term "cellular agriculture," which was coined in 2015, refers to the alternative method of producing goods that are typically produced from animals by using bioreactors rather than traditional livestock husbandry (Stephens, et al., 2018). In addition to revolutionizing the food sector, this innovative technology may contribute to future solutions for ensuring food security. Precision fermentation has become a component of cellular agriculture in this context. It is a wide notion that includes all fermentation processes that are carefully optimized using host microorganisms that are specially created to behave as "cell factories" and produce high-value functional products (Teng, et al., 2021). Even while precise fermentation has been used for decades to create a variety of chemicals, especially enzymes, it has become a viable alternative to using animals to get animal proteins in the context of cellular agriculture as cited by Marilia M. Knychala, (2024).

In the food industry, it provides environmentally friendly substitutes for conventional animal husbandry. For instance, it enables the creation of novel baby milk formulas, confections, and wholesome beverages, as well as flavorings for plant-based meats. It drives drug development innovation in the healthcare industry by facilitating personalized medicine and the synthesis of uncommon, complicated compounds. Precision fermentation is a relatively new technique for producing animal proteins for the food business, and it was once thought to be more expensive. However, ongoing efforts to optimize the process should help to overcome these obstacles. The commercial synthesis of human insulin, growth hormones, enzymes like rennet (Chymosin), and other biological products has been facilitated by precision fermentation since the challe. It is nearly the only method used to synthesize several vitamins and supplements as cited by Marilia M. Knychala, (2024).

The proteins made by precision fermentation are bioidentical to the conventional ones of animal origin, possessing the same nutritional value and sensory qualities as the comparable proteins offered by the "plant-based" sector. This presents a chance to solve issues related to food safety, animal welfare, and the environment while simultaneously meeting the rising need for protein (Mattick, 2018).

The production of animal-based food is one of the primary causes of the worldwide loss of natural ecosystems and is linked to the extinction of many species in the modern era as cited by Marilia
M. Knychala, (2024). The dramatic decline in biodiversity is also a matter for concern. Numerous studies have already demonstrated that the agriculture sector's current patterns of production and consumption cannot feed a growing population while also ensuring a future free from climate change (Ivanovich, et al., 2023). Fortunately, a number of issues with the current global manufacturing system may be resolved by precision fermentation. Precision fermentation has been demonstrated to have a better environmental impact than animal products, with some commodities using less water (Anon., 2021).

Moreover, life cycle assessment studies show that, in comparison to many traditional production methods, particularly those that use animal sources, precision fermentation can lead to much lower GHG emissions (specifically methane). In this regard, the quick implementation of precision fermentation technology in place of animal-derived products may be crucial in slowing down or perhaps stopping climate change by decreasing the probability of extreme weather and climatic events as cited by Marilia M. Knychala, (2024).

In order to prevent systems from being directly impacted by unfavorable weather conditions, precision fermentation enables production in a closed and completely controlled environment. Furthermore, by removing the necessity for animal slaughter, precision fermentation also addresses ethical issues pertaining to animal welfare and lowers the danger of antibiotic resistance and zoonotic illnesses, two serious public health problems linked to intensive animal farming (Linder, 2019).

Precision fermentation improves public health outcomes and ethical norms by creating safe, clean proteins without using animals. In contrast to animal products, precision fermentation generates less waste because microorganisms can be engineered to maximize yield and efficiency. Additionally, microorganisms can be rapidly grown and multiplied using inexpensive substrates (often by-products of other processes), resulting in faster and more efficient production (Jach, et al., 2022). Additionally, microbial fermentation wastes are frequently useful inputs for other industrial processes, lowering environmental pollution overall and promoting a circular economy (Boukid, et al., 2023).

Precision fermentation has the potential to significantly increase the efficiency of industrial food production by outperforming livestock farming in terms of cost, capacity, speed, and volume of food production. By lowering production costs via research and development, precise fermentation technologies will become more economical and accessible to a wider range of people, opening up new markets and employment opportunities as cited by Marilia M. Knychala, (2024).

3.2. Process

Precision fermentation is a cutting-edge technology enabling the production of complex proteins typically derived from animals by using engineered microorganisms. Unlike traditional fermentation, which generally produces simple molecules like alcohol, precision fermentation uses genetically modified microbes to create specific proteins by introducing target genes. These microbial "factories" can produce proteins identical to those found in milk, eggs, and meat, offering sustainable alternatives to animal-derived ingredients. Precision fermentation differs from recombinant DNA technology because it focuses on microbial cultivation for targeted protein production and its processing steps emphasize high-throughput fermentation, protein extraction, and purification. Recombinant DNA technology is a broader genetic modification process, potentially altering entire organism traits or producing a variety of proteins, not limited to fermentation. While both methods involve genetic engineering, precision fermentation is a more streamlined, protein-specific process within the broader recombinant DNA technology toolkit. The process of precision fermentation has six steps: selection of microorganisms and genetic engineering, fermentation process setup, induction of protein production, fermentation scaling and control, protein harvesting and purification, post-processing, and formulation.

I. Selection of Microorganism and Genetic Engineering

In host selection, microorganisms like *Escherichia coli* (bacteria), *Saccharomyces cerevisiae* (yeast), (Gomes et al., 2018) or filamentous fungi (Nevalainen et al., 2018) are often chosen as hosts due to their well-understood biology and ease of genetic manipulation. Each has its advantages; rapid proliferation, suitability for large-scale production, and cost-effectiveness are the advantages of selected bacteria. However, it is limited in producing complex proteins that need post-translational modifications. Yeast is capable of producing more complex proteins and performing some post-translational modifications, though slower to grow than bacteria. Fungi is often used for more complex protein needs as they can produce larger proteins and perform modifications similar to higher organisms (Knychala et al., 2024).

Genetic modification is done to make the microorganism produce animal-based proteins, genes encoding these proteins are inserted into its genome (Knychala et al., 2024). Techniques used here include; CRISPR-Cas9 or Recombinant DNA Technology, plasmid vectors, and selection and screening (Knychala et al., 2024). CRISPR allows for precise gene insertion, deletion, or editing, ensuring the microorganism's DNA directs it to produce the target protein. Plasmid vectors are often used to carry the target genes into the microorganism. They can also include regulatory elements (like promoters) to control when and how much protein is produced. After transformation, successful genetically modified strains are screened for high expression levels, stability, and efficient protein production. Selected strains are often further optimized for commercial production (Knychala et al., 2024).

II. Fermentation Process Setup

A balanced mix of nutrients is crucial for preparing culture media. The media typically contain a carbon source like glucose or glycerol as a primary energy source, and ammonium salts, or amino acids for protein synthesis. There are also minerals and trace elements; essential ions like magnesium, calcium, and potassium support enzyme function and overall cell health, and some vitamins as some microorganisms need vitamins (like biotin) to grow and function optimally (Caldwell et al., 2021).

Once the medium is ready, the selected genetically modified microorganism is inoculated into the fermentation tank. pH, temperature, and oxygen levels are carefully managed because they directly affect the organism's growth rate and protein synthesis (Caldwell et al., 2021). Initially, microorganisms grow without producing the target protein, allowing them to reach high biomass levels, setting the stage for efficient production.

III. Induction of Protein Production

Triggering protein expression and expression optimization are the two phases in this step. After sufficient growth, protein production is induced by manipulating growth conditions or adding an inducer. This can be done in several ways. Making temperature shifts is one way, as a sudden temperature change can trigger certain promoters that initiate protein synthesis. When using chemical inducers, in bacteria like E. coli, molecules like IPTG (isopropyl β-D-1- thiogalactopyranoside) can trigger gene expression of the target protein (Caldwell et al., 2021). During the optimization of expression, parameters are adjusted to maximize protein yield; Agitation ensures proper mixing, while aeration maintains adequate oxygen levels, both of which are critical for aerobic organisms, and Nutrients are often fed incrementally (fed-batch) to avoid depletion and control byproducts, which could inhibit protein production (Caldwell et al., 2021).

IV. Fermentation Scaling and Control

Scaling is done by transitioning from lab-scale to industrial-scale bioreactors, which requires ensuring that conditions are consistent across all scales. Here, a consistent growth environment is important to maintain pH, temperature, oxygen transfer, and nutrient concentrations (Caldwell et al., 2021). A scaled-up process (often to pilot-scale) is tested and optimized before moving to full production scale to minimize yield losses and maintain consistency. Automated monitoring and control is done. Parameters like pH, oxygen, CO_2 levels, and biomass concentration are continuously monitored with advanced sensors. Data-driven adjustments are done for feeding, aeration, or temperature to maintain ideal conditions, improving consistency and efficiency (Caldwell et al., 2021).

V. Protein Harvesting and Purification

After fermentation, the target protein is separated from the cells or culture medium. Then cell lysis (for Intracellular Proteins) is done which means mechanical, chemical, or enzymatic methods are used to break cells and release proteins. The choice depends on the cell type and the sensitivity of the protein (Caldwell et al., 2021). Filtration and centrifugation are the common methods to remove cell debris, concentrate the protein, and simplify downstream purification (Caldwell et al., 2021).

Multiple stages of purification may be needed depending on the protein's complexity and purity requirements; Techniques like affinity, ion-exchange, and size-exclusion chromatography help isolate the target protein based on its chemical properties (Caldwell et al., 2021). Ultrafiltration and diafiltration methods further concentrate the protein and remove smaller impurities. Final polishing removes any remaining contaminants, such as endotoxins, to achieve purity of food- or pharmaceutical-grade (Caldwell et al., 2021).

VI. Post-Processing and Formulation

Depending on the protein's application, it may need stabilization to maintain structure and functionality. Freeze-drying (Lyophilization) is commonly used to dry proteins while preserving their structure. Spray drying is often chosen for larger batches; it produces a stable powder form for easy storage and distribution. The protein can be incorporated into various products. Formulation may involve the addition of stabilizers or preservatives to increase shelf life and maintain quality and product-specific customization that is done depending on the end use, proteins might be blended with other ingredients to create meat analogs, dairy alternatives, or supplements.

3.3. Current Uses

Production of alternative proteins is one of the common uses of the technology of precision fermentation. It is used to produce recombinant milk and egg proteins, which are entering the market as sustainable alternatives to traditional animal-based proteins (Nielsen et al., 2023). This technology is also being applied to create meat substitute analog components, targeting the use of these proteins as additives in various food products (Nielsen et al., 2023).

Another use of precision fermentation is as a designer enzyme for food processing; it enables the production of tailor-made enzymes with improved catalytic properties, enhancing efficiency and adaptability in food processing (Boukid et al., 2023). The use of synthetic biology and gene editing techniques has refined the production of these designer enzymes, although scale-up remains a challenge (Boukid et al., 2023).

Sustainable food ingredients are produced using precision fermentation. Advances in strain engineering and downstream processing are crucial for producing fermentation-derived food ingredients such as proteins, fats, and oligosaccharides (Augustin et al., 2023). The integration of synthetic biology with fermentation processes supports the development of sustainable food ingredients, reducing reliance on traditional agricultural methods (Augustin et al., 2023).

Precision fermentation is exploring the use of electro-synthesized acetate as an alternative carbon source, which can reduce production costs and improve market price stability (Crandall et al., 2023). This approach helps preserve arable land and offers a sustainable method for producing food and chemicals (Crandall et al., 2023).

4. Application of Precision Fermentation as an Alternative for Animal-based Food Production

Due to the densely increasing population over the past decades production and consumption of meat, egg, and milk have increased, as they contain high amounts of bioavailable proteins with essential amino acids compared to most plant-derived proteins. To meet this demand, the amount of protein that must be produced in 2050 must be 80% higher than the current level of production (Boland & Hill 2020). Precision fermentation offers a sustainable and ethical alternative to traditional food production by precisely controlling the fermentation conditions to produce identical or closely similar versions of animal-derived proteins since it can yield a variety of proteins, fats, and other food ingredients that replicate the taste and functionality of animal-based products. Milk whey and egg proteins are extremely challenging to replace by plant proteins due to their unique structural characteristics which furnish them with many functional properties (Dilek et al., 2023).

4.1. Types of proteins

4.1.1. Meat

proteins

Collagen

Protein

Precision fermentation represents a disruptive platform in food technology; the development of animal-derived proteins like collagen is, therefore, of special relevance. Some obvious merits such techniques represent include safety, sustainability, and functionality. Collagen is the most important structural protein in animal bodies, accounting for approximately 30% of the animal's total protein content (Liu et al., 2024). Traditionally, collagen is derived from the skin of fish, pig tissues, and cow hides; due to its stability, gelation, and emulsification functional properties, it has found wide applications in the food, medicine, and cosmetic industries. There is an urgent need for alternative means against the negative impacts of environmental degradation, immunogenic reactions, and pathogenic transmissions emanating from the production of animal-derived collagens (Wang et al., 2023). This therefore means that because of the emulation in triple-helix shape and amino acid sequence from animal-derived collagen, production of collagen through precision fermentation by microbial systems is increasingly being carried out (Liu et al., 2024; Wang et al., 2023).

Most of the biotechnological methods of production involve genetic engineering of microbial hosts, including but not limited to Escherichia coli and Komagataella phaffii, to program them for the expression of collagen or collagen-like proteins. In most cases, such production has been done in a two-phase fermentation process whereby a high cell density is achieved during an initial batch phase, followed by the fed-batch to maximize protein expression (Liu et al., 2024). Although recombinant collagen is made of shorter polypeptide sequences and lacks several of the post- translational modifications present in native mammalian collagen, genetic engineering of recent technology has enabled the addition of enzymes like hydroxylases, which facilitate proline and lysine hydroxylation to give a structure and functionality closely similar to that of native collagen (Gopinath et al., 2022).

Companies like Geltor, Modern Meadow, Jellatech, and Evonik have been reporting impressive development milestones with precision fermentation technology in order to scale up the production of recombinant collagen. Their portfolios comprise collagen proteins for nutraceutical, cosmetic, and food product applications. These companies manufacture collagen by precision fermentation that can be reproduced with quality and lower allergenicity compared to its conventional animal- derived form (Liu et al., 2024). Other sustainability issues are addressed in the manufacture of PF collagen in that there would not be a reliance on animals, reducing the impact on land and water use to minimize carbon emissions. There is negation of the need for animal sources and their incidental contribution to land and water use and, consequently, carbon emissions Wang et al. (2023). Moreover, the very neutral sensory properties of PF collagen make it extremely suitable for incorporation into food products because this presents an alternative from non-animal sources to traditional gelatin in products such as gummies, desserts, and dietary supplements (Liu et al., 2024).

Precision fermentation is another route to collagen production, responding to increased consumer demand in the food industry for ingredients that are truly sustainable and animal-free. Considering outstanding challenges, such as structural complexity reached with animal-derived collagen, there is definitely room for optimism from both commercial scaling and technological improvement regarding the precision of fermentation for wider application in the reduction of animal agriculture and supporting a shift toward a more sustainable food system (Gopinath et al., 2022; Liu et al., 2024).

Hemoglobin Protein

Hemoglobin is a very important hemoprotein with an incorporated heme prosthetic group—an iron-centered complex bonded to an organic porphyrin ring. The most well-known hemoprotein, hemoglobin, plays a paramount role in oxygen transport, pH control, and erythrocyte metabolism in vertebrates (Li et al., 2022). Besides its physiological value, this protein has numerous applications in medicine and the food industry. Medically, recombinant human hemoglobin has been studied as an acellular oxygen carrier due to the high demand for blood transfusions throughout the world. In the food industry, hemoglobin is an active ingredient that can be used as an iron supplement; it also acts as a natural colorant or flavor enhancer because of its red pigmentation (Li et al., 2022; Chen et al., 2022). Animals have been the traditional source for this protein. Issues of sustainability, infection risk, and batch-to-batch unpredictability are making animal sourcing increasingly difficult. In contrast to its equivalents derived from animals, precise fermentation allows for the generation of hemoglobin through microbial expression systems that are both less harmful to the environment and of more uniform quality (Smith et al., 2024).

Precision fermentation utilizes a genetically modified microbial host, such as but not limited to E. coli, K. phaffii, and Saccharomyces cerevisiae, for producing hemoglobin in a controlled environment. However, balanced expression of its α- and β-globin subunits and sufficient heme biosynthesis for functional protein generation have various limitations. E. coli has shown promise for hemoglobin production by expressing a heterologous heme transport system that enables sufficient heme delivery, whereas optimization of the expression of the genes encoding for heme synthesis is still the primary focus of investigation (Li et al., 2022). Despite yeasts like K. pneumoniae and S. cerevisiae producing much lower titers other studies involving cerevisiae have also made progress with intracellular hemoglobin production, quoting yields as high as 18% of total yeast protein—a threshold at which scalable production is possible (Chen et al., 2022; Liu et al., 2023). Recombinant hemoglobin represents just one vanguard of the new demand from consumers for ingredients with sustainable and animal-free origins, as its utility extends further to functional and nutritional enhancement employing food coloring. For example, hemoglobin itself is a natural iron fortificant that could enrich nutritional profiles without giving a metallic taste associated with synthetic supplements of iron (Smith et al., 2024) This coming technology answers the call to action due to a global challenge brought by iron deficiency anemia through its potential to have better public health in improving bioavailable iron in the diet. While precision fermentation is one promising route towards efficiently producing iron-fortified food products, challenges persist due to complexity and high costs from the purification processes that may limit scalability and accessibility.

In addition, recombinant hemoglobin has some very desirable textural and coloring effects in plant-based meat analogs, therefore making an authentic and visually appealing product available to the consumer (Li et al., 2022; Chen et al., 2022). Precision fermentation, therefore, enables the production of hemoglobin to a high reproducibility, safety, and lower environmental footprint compared to more traditional methods; it is a transformational approach to the sustainable production of hemoproteins.

Myoglobin Protein

Myoglobin is a cytoplasmic hemoprotein, primarily found in cardiac myocytes and oxidative skeletal muscle fibers and functions and structures similar to hemoglobin. It reversibly binds to oxygen facilitating oxygen transport from red blood cells to mitochondria during increased metabolic activities such as intense physical activities, supporting muscle endurance and performance, and acting as an oxygen reservoir during hypoxic or anoxic conditions due to its high affinity for oxygen. Myoglobin plays a crucial role in the sensory qualities of meat, related to the red tone and metallic flavor (Ordway, 2004).

Recently many food processing companies have achieved myoglobin which is produced by precision fermentation that replicates the functional and sensory properties of animal-derived myoglobin and incorporates the protein in plant-based and lab-grown meat products since it provides realistic meat color and flavor that mimics traditional meat, in terms of the distinctive red color and umami taste in real meat. According to Varadarajan, et. al (1985) myoglobin is produced using E. coli on a gram scale quantity. After using yeast platforms, the most effective production has been achieved by K. phaffii, and results were further improved by boosting heme biosynthesis and inhibiting heme degradation (Yu et al., 2023). Soy hemoglobin and bovine myoglobin expressed in K. phaffii are utilized by companies like Impossible Foods Inc. (Redwood City, CA, USA) and Motif FoodWorks (Boston, MA, USA) to produce meat analogs such as Impossible Burger and HEMAMI, respectively (Marilia et al., 2024). Optimizing fermentation conditions in genetically modified yeast strains also resulted in higher yields (Zhang et al., 2021).

4.1.2. Dairy proteins

Precision fermentation is used as a sustainable and innovative alternative to produce dairy proteins such as whey, casein, and different bioactive proteins like lactoferrin, without the use of traditional milking animals. It answers many emerging concerns related to traditional animal-based dairy farming and production like environmental sustainability, resource-intensive husbandry practices, methane-like greenhouse gas emissions, animal welfare, and satisfying the global demand for dairy products with the increasing global population. Here controlled growth of microorganisms and microbial enzymatic processes are used to replicate major two dairy proteins and also to produce bioactive proteins that enhance the nutritional profile and health benefits for consumers. The protein fraction of milk consists of 80% caseins and 20 % whey proteins of its 3-3.9% of total protein composition, comprising all the essential amino acids.

Whey protein is comprised of 52 % b-lactoglobulin (BLG), 17% a-lactalbumin (aLA), 12% glycomacropeptides, 10% immunoglobulins, 5% serum albumin, 1.5% lactoferrin, and 2.5% other proteins. The food protein ingredient market is dominated by both dairy (mainly by whey) and egg proteins, together owning 40-70 % of the revenue in the animal protein ingredients market (Dilek et al., 2023). Due to the valuable and versatile functional and nutritional qualities of the whey, they are largely used in meat and milk products, low fat products, confectionery, bakery, sport formula and infant formulas.

β-lactoglobulin (BLG), is the main protein found in ruminant milk whey, making up 10-15 % of total milk proteins, which is not present in human milk (Aich et al., 2015). Bovine BLG consists of 162 amino acid residues and has 18.4 kDa of weight. BLG monomer has one free cysteine and two disulfide bonds in a calyx-shaped globular protein. Due to its amino acid sequence and structural properties, it is known as a lipocalin which is a diverse transport protein that binds with ligands such as lipids, bilins, and retinoids. The fungal host T. reesei was utilized as a non-animal source in the biotechnical production of the protein bovine beta-lactoglobulin (TrBLG). Protein was investigated using two different promoter systems to test the concept expression of its functions through a fungal host. TrBLG was successfully produced at a rate of 1 g/L using a 24- well plate and a bioreactor scale. This recombinant TrBLG was found to be N-glycosylated, predominantly with Man 5 glycans, exhibiting emulsification properties similar to those of corresponding bovine protein (Dilek et al., 2023). The US company Perfect Day (Berkeley, CA, USA) has developed β-lactoglobulin using T. reesei and used in ice cream products. According to the company's life cycle assessment, this method of producing whey protein has reduced the company's energy demand by 29-60%, water consumption by 96-99 %, and greenhouse gas emissions by up to 97% compared to using traditional cow's milk (Takeshi, 2024). Besides T. reesei, TrBLG can be produced by other recombinant organisms such as P. pastoris, E. coli, and
L. casei (Hazebrouck et al., 2007).

In addition to producing animal protein alternatives, precision fermentation can be used to introduce specific modifications to create altered animal protein structures, which lead to even superior functionalities by using amino acid substitutions. Cysteine residues can be used to increase the thermodynamic stability and structure of β-lactoglobulin, forming covalent inter- and intramolecular disulphide bonds between proteins network (Weidemann et al., 2020). BLG has five cysteine residues forming two disulphide bonds (Creamer et al., 2011). E. coli DH10B has also used to produce BLG variants and cysteine–modified variants can be used to demonstrate the role of disulfide bonds in BLG protein structure in food systems like films, emulsions, and gels, in processing (e.g., oxidation and aggregation) and digestion (e.g., hydrolysis and allerginity) achieving desirable structural characteristics of modified BLG produced through precision fermentation (Sarah et al., 2023). As this technology represents a significant advance in food science and technology, achieving optimal scalability is essential for its commercial viability. For instance, the BLG production level has increased from the microscale to the lab scale and now moving to the pilot scale, demonstrating the scalability and reliability of large-scale commercial production of BLG protein by Aspergillus oryzae in its process transitions (Marilia et al., 2024).

Lactoferrin is an 80-kDa protein in the transferrin family and it is found in various bodily fluids including milk. Lactoferrin's unique capacity to bind to cell surface receptors across different tissues enables plentiful health benefits such as cancer prevention, anemia, immune modulation, and acting as a natural antioxidant and antibacterial. Host strains like Saccharomyces cerevisiae and P. pastoris fed-batch cultivation are utilized for industrial bovine lactoferrin (blf) production through precision fermentation along with high-cell density fermentation (HCDF) to increase protein productivity, improve downstream yield, reduce production cost, and decrease culture volume (Trinh et al., 2023). Ark Bio Solution (Florianópolis, Brazil) uses lactoferrin produced by precision fermentation using host microorganism, K. phaffii, and AOX1 promoter, in foods and supplements while Helania (New York, NY, USA) uses the protein mainly in infant formulas. Using the AOX1 promoter in K. phaffii has successfully achieved higher production yields reaching up to 3.5 g/L of recombinant bovine lactoferrin. Usage of filamentous fungi such as Aspergillus awamori allows a remarkably high yield of 2 g/L than the 1.2 g/L yield of K. phaffii in producing human lactoferrin (Jiang et al., 2008).

Numerous studies have documented the expression of α-Lactalbumin from humans, cows, and goats in different host microorganisms such as E. coli, S. cerevisiae, and K. phaffii but in very low yields, less than 1mg/L. the most successful results were achieved with human α-Lactalbumin expressed in K. phaffii regulated by the AOX1 promoter, using an alpha factor secretory signal peptide and expressing human disulfide isomerase. α-Lactalbumin is essential for lactose biosynthesis and acts as a source of essential amino acids for infant nutrition such as lysine, tryptophan, and sulfur-containing amino acids. Due to α-Lactalbumin's high protein quality indicated by digestibility, rich amino acid content, neutral flavor, and water solubility, it is ideal for many food applications. Since α-Lactalbumin is the second most prevalent whey protein in bovine milk, companies like Perfect Dairy (Berkeley, CA, USA) have been investigating its recombinant production and finally produced it by T. reesei successfully (Marilia et al., 2024).

Perfect Day has incorporated recombinant whey protein into ice cream introduced by Brave Robot and into cream cheeses made by Modern Kitchen, protein powders for sports nutrition launched by California Performance Company, and animal-free milk released by Tomorrow Farms. EVERY Company has likewise launched various products using animal-free soluble egg-white proteins such as plant-based patties, protein bars, protein water, cookie baking mix, carbonated drinks, kombucha, and more. According to the United States regulations, Perfect Day's BLG cannot be labeled as "BLG", it should be labeled as "non-animal whey protein", despite its nutritional equivalence to animal-derived whey protein (Meyer et al., 2023).

Caseins have diverse biological functions such as binding metal ions, facilitating absorption of essential nutrients, and acting as precursors of bioactive peptides which regulate many significant physiological functions, making up approximately 80% of the protein content in cow's milk. Better Dairy (London, Great Britain), Change Foods (San Francisco, CA, USA), and Eden Brew (Sydney, Australia) have produced caseins expressed through K. phaffii. Perfect Day, New Culture (Berkeley, CA, USA), and Foodtech Company already use recombinant casein produced by precision fermentation using Trichoderma reesei in dairy products like mozzarella cheese. The resulting casein through precision fermentation has been reported to enhance the protein content and texture of plant-based cheese alternatives, making them more similar to their dairy counterparts. Some companies are still investigating the production of subtypes of casein such as αS1-casein, αS2-casein, β-casein and κ-casein (Marilia et al., 2024).

4.1.3. Egg white proteins

Raw eggs are a good protein source, containing approximately 12.5-13% protein. Ovalbumin is the most abundant protein in egg white, making up half (54%) of protein found in egg white while other proteins like ovofransferrin, ovomucoid, ovomucin, and, lysozyme contribute 12%, 11%, 3.5%, and 3.5% respectively. Since Ovalbumin is a complete protein, it includes all essential amino acids possessing many valuable functional properties, including foaming, emulsifying, and gelling, which are already applied industrially in cheese, confectionary production, and various other products (Małecki et al. 2021). The significant environmental impact of poultry farming and concerns over animal welfare have sparked interest in developing egg proteins through precision fermentation for the food sector. Recently this technology has been attracting global attention.

Since the 1970s, there have been efforts to express egg white proteins recombinantly, particularly chicken ovalbumin in E. coli, S. cerevisiae, and K. phaffii. While expression levels in E. coli, and

S. cerevisiae resulted in lower yields (few mg/L) and lacked concrete data on productivity, K. phaffii has resulted in higher yields by employing yeast with mutant forms of albumin fusing with other proteins, using appropriate promoters (AOX1 or a mutated AOX2 promoter), using specific yeast strains, medium pH optimization, optimal methanol and oxygen feeding and employing high cell density fermentation methods. More recently, Yang and team achieved an impressive yield of

5.45 g/L for quail ovalbumin using *K. phaffii* (Yang et al., 2009). Several companies are already developing the production of ovalbumin and other egg proteins using precision fermentation. Recent advancements include Trichoderma reesei achieving an ovalbumin expression titer of 2 g/L, while Protein Brewery has demonstrated ovalbumin expression in Aspergillus niger. EVERY Company has successfully expressed recombinant egg-white protein using Komagataella phaffii, previously known as Pichia pastoris through precision fermentation providing the same baking and cooking properties as traditional egg whites. (Aro et al., 2023).

As in the TrBLG, recombinant hen ovalbumin (TrOVA) is also produced by the fungal host Trichoderma reesei following the same method. The production level of TrOVA was 2 g/L. After producing in 24 well plate and bioreactor scale, the protein was further purified and characterized and functional properties were tested with corresponding hen ovalbumin. This recombinant TrOVA protein showed excellent foaming and heat-induced gelling properties but the strength of the gel was lower than with hen ovalbumin mainly due to the presence of other host proteins and partial degradation of TrOVA (Dilek et al., 2023).

Ovomucoid, which was recently developed by precision fermentation is a glycoprotein found in egg whites, making up nearly 11% of the total protein content. It prevents the enzyme trypsin from degrading egg white proteins, acting as a trypsin inhibitor and its defensive function contributes to the egg's natural preservation against bacteria. This has been studied for its tumor-suppressive properties, enhancing the use of an anticancer agent. Ovomucoid is also recognized as a major allergen in eggs, causing egg allergies often. Every company (Daly City, CA, USA) has produced ovomucoid expressed in K. phaffii to use in foods and supplements (Marilia et al., 2024). In K. phaffii chicken ova transferrin, the second most abundant protein in egg white has also been successfully expressed, yielding 0.1 g/L. furthermore, smaller proteins such as lysozyme and avidin have been produced suing K. phaffii, with yields of 0.4 g/L and 0.33 g/L respectively (Zocchi et al., 2003).

4.2. Animal-based Food Products by Precision Fermentation

Precision fermentation is transforming the landscape of animal-based product production by providing innovative alternatives that closely mimic traditional dairy, egg and meat products without the use of animals. This technology harnesses microorganisms to replicate proteins and other components traditionally derived from animals, thereby addressing environmental, ethical, and health concerns associated with industrial animal farming (Fatma et.al, 2023). The precision fermentation based advanced technology leverages the use of microorganisms such as yeast, bacteria or fungi to produce proteins, fats and other components that are typically obtained from animals. By programming these microorganisms at the genetic level, scientists can precisely control production of compounds that deliver the texture, taste and functionality that is associated with the traditional animal products (Karson et.al, 2024). Precision fermentation is a tool a for food innovation and also an approach to food production which offers scalable solutions to meet the growing global demands of protein in a sustainable, ethical and health-conscious manner.

4.2.1. Meat substitutes

The advancement of precision fermentation extended to the realm of meat alternatives. While plant-based meals have gained popularity, the development of cultivated meats, grown from animal cells, has also progressed significantly. Following initial proof-of-concept in 2013, cultivated meat products received market approval in 2020, signaling a viable option for consumers seeking sustainable protein sources (Marilia et.al, 2024).

Microbes are used to synthesize specific proteins, peptides, fats, and enzymes, (Fatma et.al,2023) which are carefully selected for their unique functional or sensory roles in meat products allowing replication of texture, flavor and nutritional profiles of meat (Fatma et.al, 2023). Precision fermentation has enabled the targeted ingredients production that could benefit mimicking of meat products. The targeted production of ingredients, may mimic the complex textures, flavors, and nutritional profiles of animal-based meat. Specific proteins, fats and other compounds that are traditionally derived from animals can be synthesized by utilizing microorganisms (Ting et.al, 2021). These produced key elements that can enhance the realism of meat alternatives, can be resulted in improving the appeal to both vegetarians and flexitarians.

Soy leghemoglobin has been produced by an engineered yeast *Pichia pastoris* to give the flavor and color of animal meat to plant-based burgers (Fraser et.al, 2018). This protein closely mimics the heme found in animals, which contributes to the meaty qualities of traditional meat products, including the rich color, umami taste and juicy texture. Soy leghemoglobin is approved by Food and Drug Administration (FDA, USA) to be used in plant-based meat which provides more meaty color, juiciness and taste providing a familiar authentic meat-easting experience (Simsa et.al, 2019). 'Impossible Foods' (Redwood City, CA, USA) is a company based on production of various plant-based meat products has leveraged this engineered yeast to produce a wide variety of meat alternatives. Their products, commonly known as 'Impossible burgers', are crafted to) replicate the appearance, feel and flavor of beef products and have expanded to include alternatives for chicken, pork and sausages. Soy leghemoglobin plays a major role in making plan-based meats more versatile. For instance, Impossible foods' products get brown and caramelized when cooked, a quality essential to the authentic meat experience (Julie, 2019).

'HEMAMI' is another yeast derived heme protein which delivers an umami flavor and a meaty aroma, closely resembling traditional animal-based meat. 'Motif FoodWorks' (Boston, MA, USA), launched this heme protein through precision fermentation and is available as a meat alternative option which can be purchased as finished products. HEMAMI contributes a deeper, meat-like flavor profile that improves the authenticity of plant-based options. Motif FoodWorks synthesized bovine myoglobin by *Komagataella phaffi* often referred by its obsolete name *Pichia pastoris.* This fermentation-derived myoglobin closely mirrors the structure and function of myoglobin found in animal muscles, which is responsible for the characteristic meaty aroma and color changes during cooking (Jeannine, 2024). They have successfully used the heme protein to give the meaty aroma to various plant-based meat products such as ground meat, sausages, patties, taco fillings, etc. which can improve the consumers' sensory perception of plant-based products.

4.2.2. Dairy alternatives

Plant-based beverages are water soluble extracts of legumes, nuts, cereals and oil seeds that may have a similar appearance and consistency to milk (Rasika et.al, 2021). Still, plant-based sources of dairy products are not nutritionally equivalent to milk as per several studies (Sebastian et.al,2018, Outi et.al, 2015, Smith et.al, 2022). However, plant-based milk alternatives from soy, almond, coconut, oat and rice are produced all around the world due to the increased demand. To enhance the drinking quality of milk and other dairy products, precision fermentation is one of the methods that has been newly practiced by the dairy industry.

Considering the various types of proteins in dairy products, casein, whey proteins and lactoferrin has various functions and nutritional benefits in milk. Replication of casein, whey proteins and lactoferrin being experimented and already in production by number of companies considering the nutritional and sensory enhancement of plant-based milk products and other dairy products like cheese.

'New Culture' (San Francisco, CA, USA) is one of the several companies that have stood out in the production of bovine casein through precision fermentation in animal-free cheese production, where they have already launched mozzarella cheese with the texture, flavor and melting profile of the traditional animal-based mozzarella (New Culture, 2023). 'New Culture' uses *K. phaffii* as the host microorganism in mozzarella cheese production. Other than New Culture in the US, 'Formo' in Germany, 'Better Dairy' in UK, 'Change Foods' in the US have been using precision fermentation in production of cheese where the targeted protein has been casein.

Other than casein, beta-lactoglobulin or commonly known as milk whey proteins are targeted and produced through precision fermentation in dairy product applications by companies like 'ImagineDairy' (Haifa, Israel), 'Perfect Day' (Berkeley, CA, USA) and 'ReMilk' (Rehovot, Israel). 'Perfect Day' also uses alpha-lactalbumin protein precise fermentation in their dairy product processing. The animal-free whey proteins in 'Perfect Day' are used in fresh milk production, ice cream and frozen dairy desserts (Takeshi, 2024). While, 'ImagineDairy' uses *Aspergillus oryzae*, 'Perfect Day' uses *Trichoderma reesei* and 'ReMilk' uses *K. phaffii* as host microorganisms and is sourced through bovine sources (Marilia et.al,2024). Beta and alpha- lactoglobulin produced by precision fermentation have gained high popularity considering its high digestibility, richness in amino acids, neutral flavor profile and excellent water solubility. They have been used for a variety of food applications including beverages and infant formulas as well (Kunz et.al, 1992, Lian, 2003).

4.2.3. Egg alternatives

Egg is a universal food, which is widely consumed around the world. Egg-white is valued in the food industry due to its high bioavailability, high essential amino acid content and functional versatility, which includes properties such as foaming, gelling and emulsification (Abeyrathne et.al, 2013). Without much productivity or with lower production levels, precision fermented chicken ovalbumin in *E coli, S. cerevisiae,* or *K. phaffii*) is being produced since 1970s. However, later with more productivity quail ovalbumin production through precision fermentation was done with the use of *K. phaffii* (Shaohui et.al, 2009). The second most abundant protein in the egg white after ovalbumin, ovotransferrin was also produced using precision fermentation by the use of the same yeast but with lower production yields. Smaller amounted proteins like lysozyme, avidin are also been successfully produced using the same yeast (Jiang et.al 2008). Many companies are now applying precision fermentation to create functionally identical egg proteins, through blending them with fats, emulsifiers and water to create new products. These egg products display properties such as coagulation, emulsification, leavening and binding (Thomas et.al, 2023).

'Eggmented Reality' (Kiryat, Israel) produces ovalbumin, which is extracted from the chickens for foods requiring egg white for the production. 'Onega Bio' (Helsinki, Finland) produces ovalbumin to be used as egg-white for food products. This is produced by the use of *T. reesei.* This ovalbumin is known as 'Bioalbumen'. The Bioalbumen can be successfully used in foods to produce fluffy, foamy, firm, sticky, gooey, crispy, sweet and savory characteristics to the foods. The main applications of Bioalbumen in food products are baked goods, scrambled eggs, pasta and cookies. 'Otro' (Marousi, Belgium) uses chicken sources to produce ovalbumin, and the product is available to be used in baking and in cocktail making. The 'EVERY company' (Daly city, CA, USA) is producing ovomucoid with the use of *K. phaffii* using chicken as the source. They have two egg white protein products available; EVERY protein and EVERY EggWhite. The EVERY protein product is soluble egg white protein which can be applied in making juices, smoothies, carbonated beverages, enhanced water, coffee and tea, kombucha, shakes, bars, powders, crackers and fortified foods. The EVERY EggWhite is egg white product that is all purpose and its culinary functionality matches that of a chicken-made egg white. This protein is applied in bakery products, dry mixes, patties, protein bars, confectionery, sauces, dressings, as a binder in meat analogues, pasta and noodles and cocktails.

4.2.4. Functional food ingredients

Functional foods are defined broadly as foods that supply additional physiological benefit and/or reduce the risk of chronic disease beyond basic nutritional functions of the consumer (Solan, 2000, Health Canada 1998). Some popular examples for functional foods are tomatoes rich in lycopene, spreads containing plant sterols and eggs enriched in omega-3 fatty acids. Consumer interest in functional foods continues to rise as there is a reflecting shift towards a healthier and a more sustainable food system. Precision fermentation is an innovative approach that stans out in food technology landscape as it allows for the development of functional ingredients such as proteins, vitamins, and bioactive compounds (Shahida et.al, 2023).

Functional food ingredients derived from precision fermentation, include a range of substances that offer health advantages beyond simple nutrition. The major types of proteins that are synthesized using precision fermentation which has been used in functional food ingredient production are, Lactoferrin, ovalbumin, collagen and ovomucoid. These protein, functional food ingredients have been included in various products that are being produced in number of companies around the world.

Lactoferrin is a nutrient found in mammalian milk which has important immunological properties and is both antibacterial and antiviral (Douglas et.al, 2020). Lactoferrin protein demand is driven by its applications in infant nutrition, immunological health, sports nutrition, cosmetics and maternal and neonatal nutrition (Hao et.al, 2018). Using *K. phaffii* as the host microorganism and sourced from humans, ‘Ark Bio Solutions’ (Florianopolis, Brazil) and ‘Helaina’ (New York, NY, USA) produces lactoferrin, and manufactures foods, supplements and infant formulas. Lactoferrin is also synthesized by ‘De Novo Foodlabs’ (Durham, NC, USA) sourced by humans and producing dairy and supplements and by ‘Turtle Tree’ (Singapore) also sourced from humans and producing foods and supplements.

Type III collagen is important for the development of skin and the cardiovascular system and for maintaining the normal physiological functions of the human body (Xin et.al, 1997). Type II collagen is synthesized using precision fermentation to produce biomaterials using *K. phaffii* as the host microorganism. This is practiced by 'Meadow' (Nutley, NJ, USA) by using human sources. 'Jellatech' (Raleigh, NC, USA) is also synthesizing collagen and gelatin through precision fermentation, by using human sources. They use them as ingredients in cosmetics and foods.

Peptides derived from ovomucoid is used as ACE inhibitors which help relax veins and arteries to lower blood pressure, (Nasution, 2023) lowers blood-pressure and has antioxidant properties as well (Abeyrathne et.al, 2015). Ovomucoid is synthesized using precision fermentation from chicken sources and using *K. phaffii* as the host microorganism and is added in drinks, food supplements in the products manufactured by 'The Every Company' (Daly City, CA, USA)

4.2.5. Flavoring agents

Precision fermentation is revolutionizing the field of food technology, in creating animal-like flavors for plant-based meat alternatives. Though this process, companies like Motif FoodWorks and Impossible Foods are developing heme proteins to replicate the distinct taste and aroma of meat as explained in 4.2.1. Motif FoodWorks has a developed bovine myoglobin product called HEMAMI, which provides plant-based products a rich, savory flavor and authentic meat like qualities (Jeanine, 2024). Impossible Foods, has created soy leghemoglobin, a plant-based heme protein that lends its products, a realistic iron-rich flavor and an appealing color (Julie, 2019). Both proteins mimic the animal meat flavor properties, such as juicy and slightly metallic taste.

The technology of precision fermentation allows the production of complex proteins such as hemoglobin and myoglobin by using microorganisms like specific yeast strains. Hemoproteins, which contain a heme prosthetic group, is responsible for various biological functions, including oxygen transport and intracellular pH balance, and are essential in animal tissues, especially muscles (Koji et.al, 2014). Heme proteins are highly sought after in the food industry as they impart flavor and aroma to meat products (Li et.al, 2024). The use of *K. phaffii* is able to improve heme biosynthesis and inhibit degradation, enhancing heme production yields.

4.2.6. Precision fermentation based-fat in replacing traditional animal-based products

As discussed above, precision fermentation has replaced many animal proteins and is already in use by several food companies around the world, for their production of animal-free meat, dairy and egg products. However, precision fermentation is also used in production of fats that can be used in production of dairy and meat products. ‘Palmless’ is a palm-oil brand produced by ‘C16 Biosciences’ company in the USA, through the precision fermentation technique (Takeshi, 2024). This Palmless oil, is to be introduced into the market in the year 2024. The product can be formulated into multiple food categories starting with non-dairy ice cream and cheese. Similarly, ‘Yali Bio’ company in the USA has introduced animal fats through precision fermentation (Takeshi, 2024) which is able to enhance the plant-based meat and dairy developments. ‘Yali Bio’ has created microbe strains, initiated pilot testing and has assessed small and medium scale production feasibility of their plant-based meat products using the precision fermented animal fats and is hoping to move into the commercialization. They are aimed at developing the most consumer demanded animal-based products, butter, cheese, milk and ice cream with the use of their precision fermented ingredients.

5. Comparison between Traditional Animal-based Agriculture and Precision Fermentation

5.1. Environmental Impact

Animal Agriculture: Traditional animal agriculture is among the highest contributors of greenhouse gas emissions, deforestation, water use, and land degradation. To put it into perspective, cattle farming is one of the most polluting-one that emits methane, an extremely powerful greenhouse gas (McConnochie et al., 2006).

Precision Fermentation: This technology makes use of microorganisms for producing certain proteins or food ingredients and has a lower environmental footprint. It requires less of those resources: water, land, and energy. Furthermore, it reduces emissions drastically since production can be done in controlled factory-like environments (O’Neill, J. M., et al.,2021).

5.2. Resource Efficiency

Animal agriculture involves huge areas of land, water, and feed for food production that could otherwise have been utilized directly by human beings. The feed-to-food conversion ratios are usually very poor.

Precision Fermentation: Precision fermentation operates on inputs at an extremely high return, far smaller in scale compared with conventional farming. The process requires very small areas of land and water and can be optimized to produce target proteins, enzymes, or nutrients in a hyper- efficient manner sans large-scale agriculture (Ashkarran and Mahmoudi, 2021).

5.3. Animal Welfare

Animal Agriculture: Animals are usually crowded together in deplorable conditions, giving rise to practices highly detrimental to animal health and well-being and morally questionable.(Singer, 2009)

Precision Fermentation: Precision fermentation, since it does not need animals for the making of food ingredients, totally negates issues of animal welfare. In their stead, it utilizes yeast, fungi, or bacteria to ensure that ethics in food production are being followed.

5.4. Health and Food Safety

Animal Agriculture: Animal-based food products can easily get contaminated by bacteria such as Salmonella, E. coli, and other pathogens, which result in the spread of food-borne diseases. Highly consumed animal-based food products are also associated with a greater risk of heart problems, cancer, and other health-related issues due to their high content of saturated fat.

Precision Fermentation: This can manufacture much cleaner, more consistent, and safer products with less presence of pathogens. Further, since ingredients manufactured under sterile conditions, the risk of contamination is at a minimum.

5.5. Economic viability, scalability

Animal Agriculture: The production of livestock and dairy forms the backbone of many economies and employs millions all over the world. Traditional animal agriculture is resource-intensive due to expenses related to land, feed, water, and labor; therefore, such agriculture is found to be less scalable in many regions.(Sedlak and Von Gunten, 2011)

Precision Fermentation: Precision fermentation can be greatly scaled; production can easily be scaled up without requiring large areas of land. Besides, it opens new markets for alternative proteins that could transform the global food system to have more available and more available food.

5.6. Technological Innovation

Animal Farming: The structure of animal farming is majorly based on traditional practices evolved after many centuries. Though modern technology has upgraded some processes, for example, breeding and feed optimization, the backbone process is still labor and resource-intensive. Innovations are slow due to the complexity of livestock farming and animal husbandry practices.

Precision Fermentation: Precision fermentation is an emergent area that, taking its cue from biotechnology, genetic engineering, and automation, fine-tunes production. It enables the design of microbial strains and fine-tuning the ability to produce specific nutrients or proteins with precision. This has been one of the most innovative areas and has won quite a significant amount of investment both from tech firms and biotech startups (Knight, M., et al.,2021).

5.7. Public Perception and Consumer Acceptance

Animal Agriculture: The general public has mixed feelings regarding animal agriculture. Although it forms part of the signature of most traditional diets around the world, high growth in demand has taken place with increased awareness of environmental and ethical issues arising from factory farming, thus leading to demands for plant-based products and alternative proteins. However, some segments of the population still have strong attachments to conventional animal products.

Precision Fermentation: Precision fermentation recently received much attention, at least from environmentally engaged consumers and those interested in animal welfare issues. While consumers are eager to try lab-raised meat and fermented alternatives, challenges exist regarding consumer trust, labeling, and understanding of technologies used in these emerging niches.(Caissie et al., 2023)

5.8. Regulatory and Approval Challenges

Animal Agriculture: Conventional animal agriculture has reached maturity, with a diverse regulatory framework across most countries on food safety, animal welfare, and environmental standards. However, the industry faces increasing pressure to adapt to more stringent sustainability and welfare legislation.

Precision Fermentation: Precision fermentation exists in a very complex regulatory ecosystem, mostly concerning safety standards, labelling, and the list of approved new products. Control agencies like the FDA and EFSA will have to review the safety of new fermentation-based ingredients. This approval is sluggish and unpredictable in most countries. It is very difficult to balance innovation with consumer safety.(Xu, Yang and Yang, 2022)

5.9. Supply Chain and Infrastructure

Animal Agriculture: It is based on a huge and well-developed supply chain, everything from feed production through slaughterhouses to distribution and retail. Yet it is still quite vulnerable due to outbreaks of diseases such as avian flu or foot-and-mouth disease, besides transport and processing, which pose an environmental problem.

Precision Fermentation: The value chain for precision fermentation is still in its infancy but is growing at a fast rate. The main entities along this value chain consist of plants engaged in fermentation, companies involved in the manufacturing of biotechnology microbes or microorganisms, and distribution networks concerning alternative proteins or nutritional. As the technology matures, this supply chain will integrate more strongly and become less vulnerable to disruption by environmental factors or diseases.

5.10. Carbon Footprint

Animal Agriculture: It is an extremely carbon-intensive industry, especially in the realms of red meat and milk production. Animal agriculture contributes to huge volumes of methane produced through the process of digestion in livestock, especially ruminants, which as a greenhouse gas is extremely potent when compared to CO2. Further, carbon emissions have resulted from deforestation and changes in land use brought about by livestock farming (Naylor, R. L., et al.,2005).

Precision Fermentation: It can further reduce the carbon footprint of food production by precision fermentation. Since the process is done in a controlled environment, it tends to be less energy- intensive and emits less greenhouse gas. If powered from renewable energy sources, then the carbon footprint of fermentation-based foods would almost be zero (Choudhury, R., et al.,2020).

6. Advantages and Challenges of the Technology

6.1. Advantages

The global population is projected to reach almost 10 billion people by 2050, so the need for more sustainable methods of food production has never been as great. Traditional animal agriculture is very good at providing the necessary nutrients, but it is a source of real environmental and ethical downsides, not to mention health concerns. Precision fermentation comes out as one of the most promising variants of such technology for the production of proteins, enzymes, and other biomolecules with very minimal environmental impact and without the involvement of animals. This paper discusses the sustainability, scalability, and economic benefits of precision fermentation.

Perhaps one of the biggest reasons in investments in precision fermentation can be justified is because of its minimal environmental footprint. Compared to traditional livestock farming, precision fermentation significantly decreases the levels of GHG emission, one of the leading contributors to climatic change. A study revealed that production of proteins through precision fermentation can cut down the greenhouse gases by up to 90% compared to beef production. This is attained due to high efficiency in the processes involved and the elimination of methane emissions, which is a potent greenhouse gas produced by livestock farming.

Moreover, precision fermentation requires, by far, less land and water than animal agriculture. Studies have shown that it uses 97% less land and 99% less water compared to beef farming (Demarco et al., 2023). This could be quite revolutionary in regions that are already beset with water scarcity and land degradation. Precision fermentation is a sustainable alternative for conserving vital resources and addressing environmental challenges brought about by conventional food systems.

Precision fermentation also ensures that some of the ethical issues identified with animal agriculture are addressed. Traditional livestock farming is fraught with practices that raise serious concerns on animal welfare, including overcrowding and cruel treatment. By eliminating the involvement of animals in protein production, precision fermentation ensures a cruelty-free way of production. This meets the increasing demand by ethically conscious consumers for products that reflect humane practices (Mohr et al., 2022).

Precision fermentation confers several health benefits. Unlike traditional farming, reliant on the heavy use of antibiotics and hormones to keep animals healthy and productive, precision fermentation does not require such additives. It reduces the possibility of acquiring antibiotic resistance, which has been described by the World Health Organization as a global health threat. Moreover, products resulting from precision fermentation are less likely to carry pathogens, thereby minimizing the risk of foodborne Salmonella, E. coli, and Listeria diseases (Xu et al., 2020).

Precision fermentation presents long-term benefits from an economic point of view. While more expensive than traditional animal agriculture at present, costs are likely to decrease going forward because of advances in biotechnology. Efficiency improvements in bioreactors and economies of scale could enable precision fermentation to reach cost parity with conventionally produced animal proteins by 2030 (Greenthal et al., 2022). This cost reduction would open new avenues for the widespread adoption of precision fermentation into global food markets.

Precision fermentation's economic potential extends beyond pure cost savings. Thousands of jobs will be created, not just in biomanufacturing but also in research and development, especially in those regions which are investing heavily in technologies for sustainable food. With innovation and entrepreneurship, precision fermentation thus has the potential to spur economic growth while meeting the requirements for sustainable food production.

In addition, precision fermentation enables nutritional customization. Unlike traditional food production, which is limited by the biological constraints of animals and plants, precision fermentation allows for the personalization of nutrient content. For example, proteins can be engineered to improve quality or include specific nutrients like omega-3 fatty acids, addressing nutritional deficiencies in certain populations (Beaurepaire et al., 2021). This degree of personalization has great implications in public health, allowing for the engineering of functional foods that cater to a wide range of dietary needs.

Precision fermentation also enhances food security by reducing reliance on traditional animal agriculture. Its scalable and resilient nature makes it particularly suitable for urban and resource- constrained areas. By producing essential nutrients locally, precision fermentation can help mitigate the risks associated with climate change and supply chain disruptions (Pingali, Alinovi, & Sutton, 2005).

Another critical area in which precision fermentation excels is in food safety. Conventional meat and dairy production are often plagued by the very real risk of foodborne pathogens like E. coli, Salmonella, and Listeria through slaughter, processing, and packaging. This is highly contrasted in the case of precision fermentation, which is usually done in sterile, controlled environments that significantly reduce any contamination risks (Benevenia et al., 2022).

Another added advantage of food products developed through precision fermentation is a longer shelf life because the foods do not develop microbial contamination. This extended shelf life can help reduce food waste, which is one of the most important issues in the chain of food supply around the world. Precision fermentation thus resolves safety and waste concerns all in one go to modern challenges in food production.

Precision fermentation exhibits resource efficiency. While classical agriculture creates huge amounts of non-edible biomass-for example, bones, skin, and organs-precision fermentation is much more efficient in transforming its raw materials into the protein or nutrient being targeted. In this manner, waste from by-products is minimized, with maximum usage of resources.

Furthermore, precision fermentation can upcycle agricultural co-products such as sugar from sugarcane or corn as feedstock for microbial fermentation. This is a technological method that transforms wastes into high-value proteins to support the circular economy and contribute to more sustainable food systems (Schulte & Knuts, 2022).

Precision fermentation can design food around those particular needs, whether hypoallergenic proteins are produced for people with food allergies or nutrient-enhanced foods for those populations that have deficiencies in certain nutrients (Muñoz-Cabrejas et al., 2023).

Bioactive Compounds: Precision fermentation has opened an avenue for studies into its use within the production of bioactive compounds such as omega-3 fatty acids, antioxidants, and probiotics that present health benefits and could thus be targeted for functional foods.

Precision fermentation has a sustaining role in some of the UN SDGs, including but not limited to Zero Hunger (SDG 2), Good Health and Well-being (SDG 3), Responsible Consumption and Production (SDG 12), and Climate Action (SDG 13) (Toboso-Chavero et al., 2021).

Shrinking resources, lower environmental impact, and unlocking additional sources of high-quality protein all contribute toward the direct use of precision fermentation in building a more sustainable and resilient food system.

6.2. Challenges

Precision fermentation (PF) is a process that leverages micro-organisms to produce specific type of proteins that are highly popular for the preparation of animal protein alternatives. Despite the enhanced eco-sustainability of these Precision fermented proteins, the sector still faces a number of obstacles, including formulations, customer perception, prices, and regulatory and food safety investigations. High quantities and competitive costs are necessary for PF-derived products to compete in the food business. For a given quantity of input energy, feedstocks, and labor, more protein must be generated using new bioprocessing technology and strain development advancements (J. Lucas Eastham and Leman, 2024).

Although fermentation was once used to preserve food by producing excessive amounts of acid or alcohol (the latter's intoxicated effect was a pleasant side effect), there are still hazards associated with fermentation, especially when utilizing traditional or spontaneous fermentation. Hazardous metabolites or pathogenic microbes can contaminate the finished product and endanger the health of customers. Therefore, the use of genetic analysis would contribute to increased safety through the early identification of dangerous microorganisms. Last but not least, employing non-food biomass to create completely new food items that are safe, healthy, and enticing to customers is highly promising when genomics and synthetic biology are combined to logically design desired qualities (Teng et al., 2021).

The precision fermentation process requires highly specific technologies as it controls over metabolic pathways of micro-organisms. The process of engineering micro-organisms into complex proteins consists of gene editing, optimizing fermentation conditions as well as protein editing. One of the major challenges in precision fermentation is achieving texture, taste, and functional properties of protein. It is very difficult to reach the creaminess of dairy and fibrous texture of meat in a given fermented setup (Baldwin, 2020).

Laboratory scale fermentation can produce animal proteins successfully, but in industrial scale it faces numerous complexities in arranging volumes for processing. Bioreactors, culture conditions, and downstream processing must all be carefully optimized to maintain constant yield, quality, and efficiency throughout higher production quantities (Wilkinson & Lee, 2021). Costly raw materials, energy consumption, and the requirement for specialized equipment further contribute to the high cost of scaling. It may be difficult for precision fermentation to economically compete with conventional animal farming until these production costs are reduced (Ellis et al., 2022).

Feedstock availability and cost are also related to economic challenges. For microbial growth to be sustained, precision fermentation depends on substrates such as sugars and other organic molecules. The feasibility of this technique depends on these feedstocks' affordability and sustainability. It's still difficult to find sustainable and reasonably priced ingredients, especially if demand for these substitute proteins rises quickly (Benecke, 2021). To solve this problem and improve sustainability, it might be necessary to create new feedstocks or use waste byproducts from other sectors.

A major factor in the uptake of proteins derived from precision fermentation is regulatory obstacles. To guarantee the safety and caliber of the finished items, this technique must negotiate intricate regulatory frameworks because it incorporates genetic modification and microbial fermentation. Countries' regulations vary greatly, which makes it challenging for businesses to expand their products internationally. Additionally, public uncertainty regarding genetically modified organisms (GMOs) and lab-grown products may impede market uptake, so it is imperative to address consumer perception and acceptance (Stucki, 2022).

7. Regulatory and Legal Situation

Several regulatory and legal challenges impact the growth and market entry of food products and food ingredients created using technologies such as precision fermentation, which uses microorganisms most of the time which are genetically modified through technologies such as recombinant DNA technology. These regulatory and legal challenges can limit market access, and cause problems with complying with them for businesses that are trying to introduce novel food products derived from precision fermentation. With the rising consumer awareness on the production methods and ingredients in food, regulatory bodies have to implement more safety assessments and transparent labeling practices to ensure consumer satisfaction (Mukherjee et al., 2022). The regulation of functional foods (FFs), including those made using precision fermentation, changes widely across regions, because each region has unique legislative and regulatory systems. These frameworks are designed to ensure the safety, efficacy, and appropriate marketing of such foods, while also adapting to advancements in food technology.

In the European Union (EU), foods developed through precision fermentation also come under regulation of novel foods, Regulation (EU) 2015/2283. This regulation requires that any novel food should go through necessary testing to comply with the risk assessment by the European Food Safety Authority (EFSA) before it can be authorized for placement in the EU market (Regulation
- 2015/2283 - EN - EUR-Lex, no date). Commission Implementing Regulation (EU) 2018/456 outlines the required information for consultation requests, including confidentiality provisions and the procedural steps that business operators must follow during the consultation process for novel food products. (Implementing regulation - 2018/456 - EN - EUR-Lex, no date).If genetic engineering is involved in the production of the variety of microorganisms used for precision fermentation additional regulations concerning genetically modified food and feed (Regulation (EC) 1829/2003) can apply.

FDA also has regulations on novel food products. By GRAS notice GRN 1145 β-lactoglobulin produced from Aspergillus oryzae using precision fermentation is approved for human consumption. Japan has regulation on functional foods that include regulations in Food for Specified Health Uses (FOSHU) guidelines. In Australia and New Zealand, foods containing recombinant soybean hemoglobin/leghemoglobin, must be labeled as "genetically modified," as

FSANZ (the Australian and New Zealand Food Safety Authority) requires that when the manufacturing process involving genetically engineered organisms (Nielsen et al., 2024).

8. Future Outlook

The creation of alternative proteins by precision fermentation has shown promise and has the potential to revolutionize the global food sector. With this technology, certain proteins that resemble those in animal-based diets are produced by microorganisms like yeast, bacteria, or fungi. Proteins, enzymes, or even lipids that mimic the flavor, texture, and nutritional makeup of conventional animal products can be produced by programming these microbes with particular genes. Building on decades of technological breakthroughs, the method makes it possible to produce proteins in an economical, scalable, and sustainable manner without depending on animal husbandry (Brennan, 2020).

As a result of the anticipated growth in population, the Food and Agriculture Organization, of the United Nations projects that meat and dairy consumption will increase by 102% and 82% respectively by 2050 (FAO interim report). Precision fermentation gains increasing attention where foods have similar taste, texture, aroma, appearance, and nutritional quality as animal-derived products without any ethical and environmental cost. The global precision fermentation market was valued at USD 1.6 billion in 2022 and is expected to reach around USD 67.9 billion by 2032, growing at a compound annual growth rate of 46% during the forecast period from 2023 to 2032 (precedence report 2023).

Precision fermentation-based technologies are also of interest to a noteworthy 40% of adults. More than half of respondents are open to making significant lifestyle adjustments to improve environmental consequences, according to the study, which also shows a noteworthy inclination to modify practices in favor of more sustainable ones. The food business has a considerable market potential for this kind of innovation, as seen by the significant majority of customers (77%) who say they would be prepared to test goods developed from precision fermentation if they are aware of their advantages (Hartman Group).

Precision fermentation has a big chance of upending the market for animal proteins. In comparison to traditional meat production, the method has several advantages, such as lower greenhouse gas emissions, less water use, and less dependence on agricultural land (Mattick, 2018). One kilogram of precision- fermented protein, for example, may take a fraction of the resources needed to produce beef or dairy proteins (Shepon et al. 2016). It is anticipated that production efficiency will rise even more as technology advances, increasing consumer access to and affordability of these proteins.

Precision fermentation opens the door to functional foods enhanced with extra vitamins or amino acids by enabling proteins to be tailored to suit dietary requirements or consumer preferences. Precision-fermented proteins are already being used by businesses like Perfect Day and Impossible Foods, giving the technique a boost in the marketplace. As organizations like the U.S. Food and Drug Administration start to set rules for the transparent and safe manufacturing of these new proteins, the regulatory environment is also changing (Froggatt & Wellesley, 2019).

Precision fermentation still faces obstacles to wider use, including as high production costs and customer acceptance, despite its potential. The process needs to be made more scalable and cost- effective by technological developments. Businesses must also respond to consumer concerns around genetically modified organisms (GMOs), which are frequently a component of precision fermentation procedures (Godfray et al., 2018). Nonetheless, if these obstacles are surmountable, precision fermentation may be a feasible and sustainable substitute for traditional animal husbandry, diminishing the ecological footprint and enhancing global food security.

9. Conclusion

Precision fermentation can be used for production of proteins and other food ingredients to replicate animal-derived products by using genetically modified microorganisms. It can be used as an alternative production method to replace traditional livestock farming. Precision fermentation can solve some of the environmental challenges that traditional livestock agriculture faces such as the reduction of greenhouse gas emissions, reduction of land and water use. This shift to replace animal-derived products with their replicates, made through alternative methods such as precision fermentation, aligns with the emerging group of modern consumers who are more sensitive to the issues of animal welfare and are in search of cruelty-free dietary options. Precision fermentation eliminates the use of antibiotics and hormones common in livestock farming. Because of that it can reduce public health risks associated with antibiotic resistance and zoonotic diseases. Precision fermentation faces notable challenges, including regulatory hurdles, production scalability, and consumer acceptance. Also currently the initial production costs are higher than those of traditional agriculture.

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