

Review

Unveiling the Latest Breakthroughs in Menaquinone-7 Research through Fermentation-Based Production

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Abstract: MK-7, like other biological molecules, exists in geometric isomers, including cis and trans forms, among which only the all-trans form holds biological significance. Recent studies have drawn attention to the manifold health advantages linked to the consumption of menaquinone-7 (MK-7). Nonetheless, the scarcity of MK-7 in natural dietary sources underscores the necessity for creating dietary supplements to fulfil daily intake requisites. Obtaining MK-7 involves employing production techniques encompassing solid- or liquid-state fermentation. However, upscaling this process becomes intricate in static fermentation due to challenges in heat and mass transfer. Consequently, the bulk of research on MK-7 synthesis via fermentation has concentrated on the liquid-state approach. To this end, endeavors have been dedicated to refining MK-7 biosynthesis by exploring diverse fermentation media compositions, optimal growth conditions, and even integrating nanobiotechnology methodologies. Innovative biofilm reactors, capable of facilitating biofilm attachment on plastic composite substrates, have also emerged as a promising solution, particularly when utilizing *B. subtilis* cells. The biofilm reactors exhibit robust extracellular MK-7 secretion, effectively surmounting the hurdles posed by high aeration and agitation rates. However, a demonstration of the scalability of this technology to pilot and industrial scales is still pending. This work offers an outline of the latest advancements in MK-7 research, with a specific focus on the strides made in MK-7 production through fermentation techniques. The paramount importance of the all-trans form of MK-7 is underscored, accentuating its role in enhancing human well-being. The ramifications of this work hold the potential to pave the way for novel strategies to amplify MK-7 production and formulate products with an optimized MK-7 profile, thereby promising avenues for enhancing human health and nutrition.

Keywords: menaquinone-7 (MK-7); health benefits; fermentation-based production; biofilm reactors; optimization of MK-7 biosynthesis



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1. Introduction

The vitamin K series encompasses a group of closely related fat-soluble compounds essential for human nutrition. Among these compounds are vitamin K1, also known as phyloquinone (PK), and vitamin K2, referred to as menaquinones (MKs) [1]. PK and MKs share a common 2-methyl-1,4-naphthoquinone core, but they vary in the structure of the isoprenoid group attached to the 3-position in each form [2]. PK is an individual molecule widely present in the plant and algal chloroplasts involved in photosynthesis [3]. Thus, it is readily available in numerous dietary sources such as green vegetables (e.g., iceberg lettuce, broccoli, and spinach), vegetable-oil-derived products (e.g., margarines, salad dressings, and spreads), and plant oils (e.g., cottonseed, olive, soybean and canola) [4,5]. On the other hand, MKs form a distinct group of molecules characterized by diverse side chains, designated as MK-n (where 'n' typically ranges from four to fourteen, indicating the number of repeating isoprene units). These MKs are primarily derived from microbial sources. Their crucial role lies in serving as electron acceptors within the electron transport

system. Consequently, MKs are found in specific dairy, animal, and fermented products, albeit in relatively small quantities [6,7].

MKs play key roles in vital metabolic pathways, such as promoting bone mineralization, and preventing arterial calcification, which are major health benefits associated with an adequate intake of MKs [8–10]. Furthermore, cutting-edge studies have uncovered the promising prospect of incorporating MKs into one's diet to mitigate the risk of various illnesses and globally significant diseases. These potential benefits include reducing the susceptibility to Parkinson's disease, type 2 diabetes mellitus, cancer, neurological disorders, immune disorders, chronic kidney disease, and obesity. Additionally, MK consumption has shown potential in enhancing the recovery or even prevention of coronavirus disease 2019 [11]. Nonetheless, the scarcity of MK-7 in natural dietary sources underscores the necessity for creating dietary supplements to fulfil the daily intake requirements. This work, therefore, offers an outline of the latest advancements in MK-7 research, with a specific focus on the strides made in MK-7 production through fermentation techniques.

2. Biological Activity of MK-7 Isomers

Among all MKs, MK-7 stands out for its long half-life in plasma and outstanding bioavailability, contributing to its superior therapeutic value [12,13]. However, MK-7's dietary sources are inadequate, with relatively low quantities found in mainstream consumer foods. This has driven the need for MK-7 dietary supplements with widespread demand complementing natural foods to meet the daily intake requirements of diverse populations.

The biological activity of MK-7 is influenced by its geometric isomers, including the *cis* and *trans* forms [14,15]. The presence of double bonds in the isoprenoid units determines the shape and, consequently, the biological activity of MK-7 [16,17]. As discussed, MK-7 exists in two primary forms: the all-*trans* form and various *cis* isomers. In the all-*trans* form, MK-7 showcases an extended arrangement of isoprenoid units, featuring double bonds in its *trans* molecular format which results in a linear shape. In contrast, the *cis* isomers of MK-7 adopt a non-linear structure which is due to the existence of double bonds in the *cis* structure, causing a bend in the molecular arrangement.

This structural disparity has a significant impact on the interaction of the *cis* MK-7 isomer with subcellular structures such as vitamin K2-dependent proteins and enzymes. Consequently, the biological activity of these subcellular components diminishes, leading to a reduction in their overall effectiveness.

Studies have shown that the *cis* forms of MK-7 exhibit less than 1% of the biological activity compared to the *trans* form [18,19]. Therefore, the isomeric composition of MK-7 preparations is a critical factor to consider in manufacturing MK-7-based dietary supplements.

The synthesis of MK-7 can be achieved through two main methods: natural fermentation or chemical reactions. In both cases, the manufacturing process along with the purification techniques utilized to refine the crude reaction mixture have a significant impact on the MK-7 isomer composition [20,21]. While chemical approaches are often more cost-effective, natural synthesis is preferred by consumers. Natural synthesis or fermentation-based production, which involves the use of microorganisms for industrial MK-7 synthesis, not only ensures environmental sustainability but also aligns with consumer preferences to support sustainable development initiatives [22]. However, MK-7 production through fermentation poses challenges, including low vitamin yield and the need for complex downstream processing, resulting in increased production costs and a limited affordability of the product for consumers due to its high price.

3. Fermentation of MK-7

To date, countless studies have been focused on improving fermentation yield and MK-7 concentration. Researchers have achieved this by thoroughly investigating and refining various elements of the fermentation process. These elements encompass fermentation

types, such as liquid-state fermentation (LSF) and solid-state fermentation (SSF), biofilm reactors, fermentation operating conditions, and cell treatment methods.

The natural production of MK-7 is accomplished through fermentation, utilizing a diverse range of bacterial strains such as *Escherichia coli*, *Flavobacterium*, and various members of the *Bacillus* species [23,24]. Notably, *Bacillus subtilis natto* stands out as the predominant strain and is favored for manufacturing MK-7 supplements and functional food products. This preference is owed to its remarkable high yield and recognized GRAS (Generally Recognized As Safe) status.

While many studies have utilized liquid-state fermentation (LSF) for MK-7 synthesis, solid-state fermentation (SSF) techniques offer potential advantages, including lower pre-processing energy consumption, higher productivity, reduced wastewater generation, and improved product recovery compared to LSF. Moreover, SSF enables the direct use of the crude-fermented product as a food supplement, eliminating the need for vitamin extraction and reducing the processing steps and costs. The yield of MK-7 resulting from SSF processes depends on various physicochemical as well as biochemical parameters such as used microorganism type, particle size, medium components, pH levels, substrate pre-treatment, moisture content, incubation temperature, humidity, inoculum size, aeration rate, and fermentation period. These variables play crucial roles in optimizing and controlling the fermentation outcome [11,25]. Different substrates, fermentation conditions (static vs. dynamic), and genetic manipulation have also been studied for their influence on the fermentation yield. However, static fermentation presents operational challenges due to biofilm and pellicle formation by *B. subtilis natto* [26–31]. These issues hinder large-scale MK-7 production.

It was commonly assumed that bacterial-based fermentation predominantly only produces the trans structure of MK-7, and there is limited evidence in the literature on the ratio of MK-7 isomers produced via microbial metabolic pathways [14,15,17]. However, recent studies have revealed that MK-7 extracted from natural natto exhibits varying proportions of the cis and trans isomers. This observation raises concerns about the accuracy of producers' claims regarding the source of MK-7 in their dietary supplements, suggesting the potential use of chemical synthesis instead of natural natto extracts or even the presence of cis isomers due to the manufacturing processes used to produce hard tablets or capsules. The MK-7 profile of fermented MK-7-enriched functional foods remains unclear, as most studies have only focused on analyzing the MK-7 composition of dietary supplements obtained from natural and chemical production methods. Further research is needed to explore the MK-7 composition in functional foods enriched with fermented MK-7.

Utilizing nanoparticles (NPs) has emerged as another recent trend in enhancing MK7 yield. Nanomaterials (NMs) offer promising solutions to address these limitations in MK-7 fermentation. These NMs, with structural components ranging from 1 to 100 nm in size, possess unique biological and physicochemical characteristics, making them ideal for various applications [7,32–34]. Among NMs, NPs are particularly relevant to MK-7 fermentation and have the potential to overcome the primary challenges associated with vitamin production. In particular, iron oxide nanoparticles (IONs) have been explored for bacterial cell immobilization, leading to increased MK-7 yield and improved metabolic productivity of the cells. Moreover, the superparamagnetic properties of IONs allow for cell separation and dispersion using an external magnetic field, leading to reduced downstream purification steps and facilitating process intensification. Consequently, employing IONs for bacterial cell immobilization could present an innovative solution to overcome challenges in large-scale MK-7 production.

In our past studies, we conducted the first assessment of the influence of uncoated iron oxide nanoparticles (IONs) on microbial growth and the isomer composition of MK-7 achieved through fermentation. The immobilization of *B. subtilis natto* cells led to an enhanced fermentation process, resulting in a 1.6-fold increase in the yield of the bioactive isomers compared to free cells when the optimal NP concentration was used [7,34,35].

However, we also observed that while naked IONs positively affected MK-7 production, they displayed low physicochemical stability and exhibited toxic effects on microbial cells, hindering bacterial growth. This is a significant concern, especially when considering the metabolic efficiency and viability of bacterial cells for effective cell recycling using magnetic separation.

In tackling these challenges, our research is dedicated to the application of biocompatible coatings, such as amino acids, to address the unfavorable properties of IONs. Amino acids exhibit excellent coating properties due to their biocompatibility, chemical stability, and surface activity. Many amino acids are deemed safe for human consumption and have already been utilized as biocompatible coatings in dietary supplements. Among the viable coating choices, L-Lys stands out as particularly advantageous. It enhances the interactions of nanoparticles (NPs) with bacterial cell membranes through its amine functional groups, thus promoting greater surface associations. Another amine-functionalized coating, known as APTES, shares similar properties with L-Lys but also provides additional benefits. APTES helps prevent NP oxidation and preserves their crystalline structure. Moreover, IONs coated with APTES significantly reduce biofilm formation without compromising cell viability and growth.

Based on previous studies, IONs, especially those with biocompatible coatings, have proven valuable in enhancing MK-7 production and yield during large-scale fermentation. In addition to the above advancements in MK-7 fermentation, biofilm reactors have also emerged as a promising new technology, offering an alternative to traditional SSF and LSF techniques. These biofilm reactors show great potential in facilitating enhanced MK-7 production without the limitations associated with static fermentation processes, which will be discussed in the next section [26].

4. Biofilm Reactors

Biofilms are formed when microorganisms create multicellular communities through colonization on a suitable surface. The structure and physiology of biofilms depend on the microbes and the surface environment [36]. Planktonic cells, when exposed to certain physiochemical signals, initiate contacts with surfaces and other cells, leading to the formation of microcolonies. These microcolonies then undergo developmental signals, differentiating into pillar-type structures surrounded by an extracellular polymeric substance containing fluid-filled channels. It should be noted that these transformations involve significant changes in gene expression to achieve mature biofilm formation [37].

While biofilm formations can have various applications, they are often undesirable in certain contexts, as microorganisms in mature biofilms are more resistant to inactivation compared to planktonic cells. This has been a concern for food processors and medical device manufacturers [38]. Additionally, interactions and synergy between different strains in a mixed-species biofilm matrix may confer increased resistance to chemicals, antibiotics, and predators [39], shedding light on the reasons behind biofilm formation by microorganisms.

In the context of fermentation, biofilms offer a natural way of immobilizing cells that enhances productivity and process stability. The formation of biofilm allows microorganisms to utilize nutrients and produce metabolites more efficiently. In response to environmental stress, some planktonic cells may adapt to biofilm formation as a survival strategy [37]. It has been observed that the production of MK-7 by *B. subtilis natto* is closely associated with the formation of pellicles and biofilms, which poses operational challenges in static fermentation. Biofilm reactors, can sustain mature biofilm formations under relatively high aeration and agitation rates, making them a cutting-edge technology to replace traditional suspended-cell or static-state bioreactors for MK-7 production [27].

Different *Bacillus* strains, including *B. licheniformis*, *B. subtilis*, and *B. amyloliquifaciens*, were previously investigated for their ability to form mature biofilm structures and MK-7 biosynthesis using glycerol and/or glucose-based media [27–31]. *B. subtilis natto* emerged as the most promising candidate, producing the maximum concentration of 35.5 mg/L of

MK-7. Moreover, four different types of Plastic Composite Support (PCS) compositions were evaluated for hosting *B. subtilis natto* biofilm formations and maturation [27]. Eventually, the combination of 50% polypropylene, 5% soybean flour, 40% soybean hulls, 5% yeast extract, and minor salts was chosen to fabricate the PCS and construct the biofilm reactors as shown in Figure 1.



Figure 1. PCS grid setup in bioreactor.

During further investigations, the growth conditions in the biofilm reactors were carefully optimized. The optimum conditions were found to be a temperature of 35 °C, a 200 rpm agitation speed, and a pH of 6.58. As expected, the introduction of active aeration and agitation in the biofilm reactors led to a decrease in MK-7 secreted concentrations, resulting in 12.1 mg/L compared to static fermentation conditions. However, despite the reduction, MK-7 biosynthesis was still 58% higher compared to suspended-cell culture, in which the biofilm formations were absent. Following this, the composition of the glycerol-based medium was fine-tuned through optimization efforts. The suggested medium composition involved using 48.2 g/L of glycerol, 8.1 g/L of yeast extract, 13.6 g/L of soytone, and 0.06 g/L of K₂HPO₄. [31]. This composition has proven to be superior for MK-7 biosynthesis and has remained a promising choice for enhancing MK-7 production.

Glucose is also known to be an ideal carbon source for *B. subtilis* strains, providing faster metabolism outputs. By utilizing the glucose-based medium, significantly higher MK-7 concentrations in biofilm reactors were observed as compared to suspended-cell bioreactors. After optimizing the fermentation media composition in the biofilm reactors for the case of a glucose-based medium, an MK-7 concentration of 18.5 mg/L was achieved. This concentration was 237% higher than the concentrations in suspended-cell bioreactors [26].

In the batch biofilm reactors, both glycerol and glucose were effectively utilized, depleting during the fermentation process. Previous studies involving *B. subtilis natto* have also shown that depletion of simple carbon sources is a common occurrence and using fed-batch supplementations can significantly improve biomass and metabolite productions [40]. Therefore, to enhance MK-7 biosynthesis, fed-batch additions of carbon sources such as glucose or glycerol were investigated. The results showed that when the fermentation was initiated with glucose, the biofilm metabolism excelled, and fed-batch implementations with either glucose or glycerol significantly supported the metabolism of *Bacillus* cells to achieve higher MK-7 concentrations. Notably, the highest concentration of 28.7 mg/L was observed with glucose supplementation. It is worth mentioning that previous studies have demonstrated that glycerol lacked the ability to sufficiently support the biofilm to endure and utilize the added carbon sources, resulting in substrate inhibition. However, upon examining the biofilm formations on the PCS grids through SEM observations, it was shown that mature colonies existed on the surface as well as in the porous environment [27–31].

Biofilm reactors hold great potential for industrial-scale MK-7 production. However, it is important to note that biofilm reactors for MK-7 production have only been studied in benchtop settings and have not been scaled up to larger systems, making their application on a larger scale yet to be demonstrated. Scaling up biofilm bioreactors with the presence of PCS supports would require unique scale-up strategies different from those used in traditional CSTR reactors.

5. CFD Simulation in Industrial Processes

Scale-up refers to the process of increasing the size of an industrial operation from the laboratory or pilot scale to a larger production scale. It is a crucial step in the development of various industrial processes, including chemical reactions, mixing, and fluid flow systems. Achieving successful scale-up is challenging due to the complexity of physical and chemical interactions at larger scales. Computational Fluid Dynamic (CFD) simulation has emerged as a powerful tool for understanding and optimizing these processes. It is important to explore the significance of scale-up and how CFD simulation plays a vital role in improving efficiency, safety, and cost-effectiveness in industrial applications.

Scaling up a process involves transitioning from a controlled environment at the laboratory scale to larger production setups. This transition introduces several challenges that must be addressed to ensure the process remains efficient and economically viable. One of these challenges is fluid dynamics, where changes in scale can significantly alter fluid flow behavior, leading to variations in mixing, heat transfer, and mass transfer rates. Understanding these changes is essential for maintaining process efficiency and product quality. Turbulence and mixing are another important aspect; turbulent flow behavior becomes more prominent at larger scales, affecting the mixing of reactants and heat transfer. This can result in reduced reaction rates and product yield if not appropriately addressed during scale-up. Heat transfer is usually another important factor because as the process size increases, so does the potential for heat accumulation and dissipation. Ensuring adequate heat transfer becomes crucial to preventing safety hazards and maintaining the desired reaction conditions. Finally, residence time distribution is also a critical factor. Residence time distribution is often different at larger scales, impacting the reaction kinetics and product selectivity. Accurate predictions are essential for process optimization.

Computational Fluid Dynamics, or CFD simulation, is a numerical tool used to solve complex fluid flow problems. It has become an indispensable tool in various industries due to its ability to provide valuable insights into fluid dynamics, heat transfer, mixing, and other process-related phenomena. In scale-up, CFD simulation plays a vital role in addressing the challenges mentioned above and ensuring a smooth transition from laboratory- to industrial-scale processes [41].

CFD simulations enable engineers to predict the behavior of fluids in complex geometries, such as reactors, pipes, and vessels. This allows for a detailed understanding of how fluid flow patterns change with scale. Engineers can identify potential issues, such as dead zones or regions of inadequate mixing, and design improvements to optimize fluid flow at larger scales. Turbulence models in CFD simulations also can help to accurately predict the effects of turbulence on mixing and heat transfer. By employing the appropriate turbulence models, engineers can ensure that the mixing efficiency remains consistent during scale-up, avoiding unexpected variations in product quality. Moreover, CFD simulations are invaluable in analyzing heat transfer mechanisms in industrial processes. Engineers can use these simulations to design efficient cooling systems and ensure temperature control during scale-up, mitigating safety risks and maintaining optimal reaction conditions. It must also be noted that CFD simulations can accurately predict residence time distributions in reactors and other flow systems [40,41]. This information helps in optimizing reaction kinetics and improving product yield. By ensuring consistent residence time distributions at different scales, engineers can minimize product variability and increase process efficiency.

CFD simulations aid in the design and optimization of the equipment used in industrial processes. Engineers can use CFD to assess different designs, materials, and

configurations before physically building the equipment. This reduces the need for costly and time-consuming trial-and-error approaches during scale-up. In addition, CFD simulations are instrumental in assessing safety aspects, including pressure and temperature profiles, to prevent hazardous conditions. They also assist in analyzing the environmental impact of industrial processes, helping companies comply with regulations and minimize their carbon footprint. CFD simulations can save both time and money during the scale-up process. By simulating various scenarios virtually, engineers can quickly identify optimal conditions and make informed decisions without the need for extensive experimental testing.

To date, in the chemical industry, CFD simulations have been used to study various types of reactors, including fluidized beds, packed beds, and stirred tanks. By understanding the fluid dynamics, mixing, and temperature distribution in these reactors, engineers can optimize the operating conditions and ensure an efficient scale-up. In addition, CFD simulations are widely used in pharmaceutical manufacturing to analyze fluid flow, mixing, and heat transfer in mixing vessels and bioreactors [40,42,43]. This helps in ensuring a uniform distribution of reactants and controlling temperature during large-scale production. In the oil and gas industry, CFD simulations have become an essential tool to study fluid flow and heat transfer in pipelines, heat exchangers, and separation equipment. Understanding flow behavior at different scales is crucial for efficient and safe operations.

As discussed, scale-up is a critical step in the development of industrial processes, and its successful implementation requires a thorough understanding of fluid dynamics, heat transfer, and mixing at larger scales. CFD simulation has proven to be an invaluable tool in addressing the challenges of scale-up by providing detailed insights into fluid flow behavior, turbulence, and heat transfer. However, there have been no attempts to utilize CFD simulations in the fermentation of MK-7. Although promising alternatives such as biofilm bioreactors or the application of nanoparticles in developing intensified bioreactors have shown potential for MK-7 production, none of them have demonstrated scalability at either the pilot or commercial scale to date. Incorporating CFD simulations into the process can empower engineers to optimize efficiency, ensure safety, and minimize costs during the transition from laboratory- to industrial-scale operations. By leveraging CFD, the trial-and-error process can be significantly reduced, saving valuable time and resources and enabling a more reliable scale-up strategy. It is essential, however, to validate the CFD results through real experiments to ensure the scalability of novel bioreactor systems for MK-7 production. The role of CFD simulation continues to expand, providing industries with the means to achieve improved productivity, sustainability, and innovation in their processes.

6. Challenges and Future Perspectives

Extensive research has been dedicated to advancing the fermentation of MK-7, focusing on improving both its yield and concentration. This endeavor has encompassed a range of explorations, including investigations into various fermentation methods such as liquid-state fermentation, solid-state fermentation, and the implementation of biofilm reactors, all under varying operating conditions. The utilization of diverse bacterial strains, prominently led by *Bacillus subtilis natto*, has been integral to MK-7 production. While liquid-state fermentation has traditionally dominated this field, solid-state fermentation presents notable advantages, such as reduced energy consumption and the direct utilization of the produced product. However, the challenge of biofilm formation during large-scale production has become apparent, warranting consideration.

Recent insights have unveiled distinct cis and trans isomers in naturally sourced MK-7, prompting a re-evaluation of the production methods. In this context, the exploration of iron oxide nanoparticles (IONs) has emerged as a promising avenue for enhancing MK-7 synthesis. The incorporation of biocompatible materials, such as amino acids, onto these nanoparticles not only boosts their effectiveness but also addresses concerns related to toxicity. These strides, coupled with the potential presented by biofilm reactors, mark a

transformative phase in MK-7 fermentation. This signifies a shift towards refined production techniques and the amplified availability of this valuable nutrient.

Furthermore, the realm of synthetic biology and genetic engineering stands poised to propel the creation of microbial strains engineered for superior MK-7 production. Delving deeper into the metabolic pathways responsible for MK-7 biosynthesis will pinpoint key bottlenecks, allowing for precise manipulations. Leveraging omics technologies encompassing genomics, transcriptomics, proteomics, and metabolomics will provide invaluable insights into optimizing fermentation conditions and refining strain development.

Innovations within bioprocess engineering, including high-throughput screening and automated fermentation systems, are set to streamline the selection and scaling of these engineered strains. To facilitate bioreactor design and scaling, computational fluid dynamics (CFD) emerges as a valuable asset. By simulating fluid dynamics, mixing patterns, oxygen transfer, heat dissipation, nutrient distribution, and waste accumulation within bioreactors, CFD offers a platform for designing the optimal growth conditions and optimizing MK-7 production. The integration of CFD with metabolic models further allows for the dynamic optimization of both strain behavior and process conditions. Importantly, CFD minimizes the necessity for exhaustive experimentation, aids in troubleshooting unexpected outcomes, and sheds light on pivotal fermentation parameters.

7. Conclusions

This study underlines the vital significance of the all-trans isomer of MK-7 within dietary supplements, underscoring its crucial role in ensuring their biological effectiveness. The therapeutic potential of MK-7 in mitigating cardiovascular diseases, osteoporosis, and other health promoting attributes highlights its profound impact on human health. *Bacillus subtilis natto* has emerged as a pivotal strain in the industrial production of MK-7, and the ongoing endeavors are concentrated on refining fermentation processes, optimizing growth conditions, and fine-tuning medium compositions to attain heightened yields.

This work presents a comprehensive overview of potential avenues for MK-7 fermentation. Amid the diverse technologies explored, biofilm reactors exhibit encouraging promise as a viable departure from conventional static fermentation methods. They effectively surmount the inhibitory effects of vigorous aeration and agitation on MK-7 production. Notably, benchtop stirred-tank biofilm reactors that foster mature biofilm formations on plastic composite supports have showcased amplified MK-7 biosynthesis.

Nonetheless, to actualize large-scale production, it is imperative to delve deeper into the impacts of the upscaling process for biofilm reactors, both on pilot and commercial scales. By addressing these pivotal junctures, biofilm reactors stand poised to emerge as a pioneering technology in industrial MK-7 production, ushering in novel prospects for advancing human health and well-being. The ongoing exploration and application of biofilm reactors in MK-7 production augurs a promising future, addressing the escalating demand for this essential vitamin and its potentially far-reaching benefits.

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