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A SCOPING REVIEW

Whey in 3D Printing: A Scoping Review

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ABSTRACT

Introduction: The issue of dairy whey utilization remains relevant despite advancements in modern processing technologies such as membrane methods, biotechnological approaches, and preservation. Global whey production exceeds 160 million tons annually and continues to grow, necessitating new solutions within the circular economy framework. In recent years, Industry 4.0 technologies, including 3D printing (3DP), have emerged as promising tools for processing dairy by-products. However, adapting whey protein products for 3DP requires further investigation of their properties and modification methods.

Purpose: This scoping review aimed to analyze the potential and current applications of whey protein products as components of 3DP inks.

Materials and Methods: The review was conducted following PRISMA-ScR guidelines. Literature searches were performed in ScienceDirect, Scopus, and PubMed (2010–2025) using targeted keywords. VOSViewer was employed for thematic analysis of the research field.

Results: Analysis of 56 selected sources revealed that whey protein components (76% of cases involving WPI) are actively studied as ingredients for 3DP inks. Their potential applications span food production, including functional and personalized nutrition (e.g., for individuals with dysphagia), as well as biomedicine, tissue engineering, and the chemical industry. Research primarily focuses on the rheological, textural, and microstructural characteristics of 3DP materials, alongside modification methods: adjusting ink composition, pre-3DP processing (pH regulation, thermal and mechanical treatment), and post-printing techniques (drying, carbonization, microwave treatment).

Conclusion: The review confirms the promise of whey proteins in 3DP materials. To advance research, the authors recommend systematizing knowledge on key components combined with whey proteins, predictive modeling of optimal formulations based on intermolecular interactions and functional properties, and integrating other whey-derived ingredients, such as hydrolysates, into 3DP applications.

Keywords: 3D printing, whey, 3D printing inks, 3D printing ink properties

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Молочная сыворотка в 3DP: обзор предметного поля

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РИДИТОННА

Введение: Проблема утилизации молочной сыворотки остается актуальной, несмотря на развитие современных технологий переработки, таких как мембранные методы, биотехнологические подходы и консервирование. Глобальное производство сыворотки превышает 160 млн тонн в год и продолжает расти, что требует поиска новых решений в рамках концепции циркуляционной экономики. В последние годы технологии Индустрии 4.0, включая 3D-печать (3DP), привлекают внимание как перспективный инструмент для переработки побочных продуктов молочной промышленности. Однако адаптация сывороточных белковых продуктов для 3DP требует дополнительного изучения их свойств и методов модификации.

Целью настоящего обзора предметного поля стало изучение и анализ потенциала и текущего применения белковых продуктов переработки молочной сыворотки, как компонентов в составе чернил для 3DP.

Материалы и методы: Обзор выполнен в соответствии с руководством PRISMA-ScR. Поиск литературы проведен в ScienceDirect, Scopus и PubMed (2010–2025 гг.) с использованием целевых ключевых запросов. Для анализа структуры предметного поля использован VOSViewer.

Результаты: Анализ 56 отобранных источников показал, что сывороточные белковые компоненты (в 76% случаях WPI) активно исследуются в качестве ингредиентов для разработки 3DP чернил. Их потенциальное применение охватывает производство пищевых продуктов, включая функциональное и персонализированное питание (в том числе для людей с дисфагией), а также биомедицину, тканевую инженерию и химическую промышленность. Основное внимание исследователей в данном поле уделено изучению реологических, текстурных и микроструктурных характеристик разрабатываемых 3DP материалов, а также методов их модификации: изменения состава рецептур, технологической обработки до 3DP (регулирование pH, тепловая и механическая обработка) и после нее (сушка, карбонизация, СВЧ).

Выводы: Результаты обзора подтверждает перспективность применения сывороточных белков в составе материалов для 3DP. В качестве рекомендаций по развитию исследований в данном поле авторы предлагают уделить внимание систематизации накопленных знаний по ключевым компонентам в комбинации с сывороточными белками, прогностическому моделированию оптимальных комбинаций компонентов в рецептуре 3DP материалов, базируясь на их способности к межмолекулярным взаимодействиям и значимым свойствам, а также внедрению других сывороточных белковых ингредиентов, например гидролизатов, в активное использование для 3DP.

Ключевые слова: 3D-печать, молочная сыворотка, чернила для 3D-печати, свойства чернил для 3D печати

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INTRODUCTION

There are technological solutions that enable the processing of large volumes of whey, a by-product of cheese production. These include membrane technologies, preservation methods, and biotechnological approaches. However, an ever-growing population and the continuous establishment of new dairy facilities continue to drive an increase in dairy waste, keeping the issue relevant (Chourasia et al., 2022; Chaudhary et al., 2023). According to Sharma et al. (2018), global whey production exceeds 160 million tons per year and continues to grow by 1-2% annually. Mohapatra et al. (2025) also emphasize the growing challenge of dairy industry waste generation that accompanies its scaling and highlight the importance of developing a circular economy approach. Within this framework, sustainable management of by-products becomes a practical objective. In the context of developing a circular economy in food production and waste processing, Hassoun et al. (2024) highlight Industry 4.0 technologies (including 3D printing) as promising tools. Thus, to prevent a food crisis while ensuring environmental safety, the active integration of modern technologies for processing dairy by-products, particularly whey, is essential.

An effective food waste management strategy is one of the key priorities across the processing industry, which is why researchers worldwide are investigating the adaptation of by-product-based raw materials for 3DP (Carvajal-Mena et al., 2022; Uranga et al., 2024; Aït-Kaddour et al., 2024). A significant challenge in using food-based and by-product raw materials lies in their frequent failure to meet the material requirements for 3DP (Jeon et al., 2024). The key indicators determining the suitability of 3DP inks for extrusion-based printing, which, according to Carvajal-Mena et al. (2022), have demonstrated the greatest adaptability to the food industry, include rheological properties (e.g., viscosity, thixotropy, elastic modulus, consistency, and flow index) and structural-mechanical properties (e.g., adhesion, elasticity, strength). As a result, researchers in this field use additional technological operations such as pH adjustment, heat treatment, and ultrasound processing to improve these characteristics (Daffner et al., 2021a; Maiz-Fernández et al., 2022; Gong et al., 2025), as well as experiment with ink formulation and printing parameters (Feng et al., 2018; Thakur et al., 2023), which may impact the final product. The growing interest in 3DP and its synergy with food raw materials has led to numerous review publications. These are typically structured either from the

perspective of application areas toward printable materials, or vice versa-from ink raw materials toward potential 3DP applications.

Several reviews focus on the use of food-based ink components for medical purposes (Sun et al., 2022; Taneja et al., 2022; Gogoi et al., 2024; Ghobadi et al., 2025). For instance, Sun et al. (2022) analyze the role of 3DP in skin wound regeneration and materials (e.g., alginate, chitosan, gelatin, polylactic acid) that can be used for this purpose, highlighting advantages such as patient-specific customization, rapid and easy manufacturing, and design flexibility. Ghobadi et al. (2025) highlight chitosan-based 3DP inks for skin wound healing as they are biodegradable, non-toxic, and possess antimicrobial activity. Other studies describe hydrogels of various origins, including natural sources, as promising 3DP inks in tissue engineering and bioscaffold production (Taneja et al., 2022; Tamo et al., 2024; Gogoi et al., 2024).

Another group of reviews focuses on the current use of 3DP in the food industry for the production of edible products, including functional foods (Liu et al., 2017; Ma & Zhang, 2022; Y. Liu et al., 2024). Ma and Zhang (2022) investigate 3DP as a tool for personalized nutrition and for modifying the sensory attributes of traditional foods, and propose a roadmap for scaling personalized food production. Y. Liu et al. (2024) discuss the key properties required of materials for 3DP and consider their potential in the production of baby food, space food, and specialized nutrition, including products for individuals with dysphagia — a topic also addressed by Z. Liu et al. (2024). Earlier, Liu et al. (2017) systematized the factors affecting the accuracy and quality of 3DP, many of which depend on the properties of the materials used. Mu et al. (2021) focus on biocompatible protein-based 3DP inks (collagen, silk, keratin; soy, egg, whey proteins), which is crucial for medical and bioengineering applications, as well as their high strength, specific rheological behavior, and hierarchical molecular assembly, which may have both positive and negative effects on the printed product. Despite a detailed analysis of the technical aspects of 3DP and specific proteins, such as keratin and fibrinogen, the authors group food proteins and describe their properties collectively. However, each protein possesses unique characteristics that require separate analysis in the context of 3DP suitability and performance. Similar grouping-based approaches were adopted by Y. Wang et al. (2024) for proteins, Feng et al. (2024) for recombinant food gels, and Jeon et al. (2024) for animal-based raw materials. However, no detailed analysis

of whey protein components (concentrates, isolates, hydrolysates, etc.) was found in such reviews, unlike for starches, which were addressed separately by Rong et al. (2023) and Chen et al. (2024).

In this regard, the present review aims to investigate and analyze the potential and current applications of whey protein products as components in 3DP inks. To achieve this aim, the following research questions were formulated:

RQ1: What specific research tasks are addressed in this field, and which methods are employed?

RQ2: Which whey-derived protein products are used in 3DP ink formulations?

RQ3: What are the application domains for inks containing whey protein components?

MATERIALS AND METHODS

Transparency Statement

The present scoping review was conducted in accordance with the PRISMA-ScR guidelines (Tricco et al., 2018). The authors confirm that the study complied with the protocol and search strategy, and any deviations are described in detail in the methodology.

Search Strategies

Database

The literature review was conducted using the Science-Direct, Scopus, and PubMed databases. ScienceDirect and Scopus databases include reliable publications with ensured peer-review, they have advanced filtering features for efficient search, and broad thematic coverage due to its focus on specialized journals in engineering, biomaterials, and food technology. PubMed was selected because its sources can complement the scoping review with materials reflecting the medical aspect of whey use in 3D printing. Articles were sorted by relevance during the search. The publication date range was restricted to 2010–2025.

2010 was chosen as the starting point due to several key considerations. As described by Su and Al'Aref (2018) in their historical overview of 3D printing (3DP), the expiration of the Stratasys patent for layer-by-layer object creation in 2005 led to the launch of two projects aimed at developing and distributing open-source 3D printer designs,

and by 2010, the models and technologies developed during the 2005–2010 period were simplified, marking a shift from the narrow application of additive technologies to their broader integration into medicine and the food industry. To identify studies aligned with the research concept, the following key search queries were used:

- 1. milk "whey" for "3D printing" / milk AND whey AND "3D printing"
- 2. "whey" for "3D printing" inks" / whey AND "3D printing" AND inks

Inclusion and Exclusion Criteria

The review included both empirical and review articles, as well as book chapters or monographs. No geographic limitations were imposed, but all included publications were in English and available in full-text format. Sources were selected based on their relevance to the review's research questions and overall concept, which, along with other selection criteria, are detailed in Table 1.

Protocol Deviations

During the sources screening identified via the first search query, duplication of information was observed between empirical research articles and review publications. Consequently, the exclusion criteria for the review were amended under the "Source Type" category. The reviewers decided to include only empirical research articles, as review articles and book or monograph chapters frequently duplicated findings from already included empirical studies or contained content that did not align with the review's context and conceptual framework.

Data Extraction

For preliminary structured annotation, the following parameters were used: authors' country of affiliation, source link, year of publication, paper title, research hypothesis, main research objectives, key findings, and conclusions. Data were extracted with the assistance of the artificial intelligence tool ChatGPT-4o.

The initial search yielded 895 sources. During the identification of the first 200 articles from the first query in ScienceDirect, a gradual decrease in relevance was observed. As a result, the reviewers decided to finish the analysis of that query and proceed with the second one.

Table 1 Inclusion and Exclusion Criteria for Sources in the Review

Criterion	Inclusion	Exclusion	Justification
Context	Studies involving the use of whey in 3DP, including its use as a whole or its components (proteins, lactose, minerals), their processing, modification, or incorporation into 3DP materials.	Studies not related to the use of whey or not involving its processing and use for 3DP, including reviews where whey is only mentioned in general.	Such criteria ensure focus on studies involving whey as a raw material or ingredient in 3DP to identify relevant strategies and assess technological potential.
Concept	Studies that analyze whey-based protein ingredients used in 3DP inks and assess their functional or structural role in various applications.	Studies that do not address whey proteins or do not consider them as components of 3DP inks.	Such criteria enable a detailed analysis of whey-based proteins, specifically in the context of 3D printing, rather than in general food applications.
Language	English.	Any other languages.	English is the dominant language of global scientific communication, ensuring access to up-to-date and peer-reviewed data.
Source type	Review and empirical papers, book chapters, and monographs.	Conference abstracts, non-peer- reviewed theses, technical reports, patents, and other non-reviewed materials.	Limiting the review to peer-reviewed publications ensures the reliability of the sources included.
Publication status	Published full-text versions are accessible online.	All sources with only an abstract are available or not published online.	Such criteria ensure full access to the necessary methodological and analytical information for adequate inclusion and comparison.

In total, 604 sources from both queries and three databases were screened. Of these, 552 were excluded for the following reasons: 35 lacked open-access full-text versions, 32 was a duplicate, 207 did not meet the inclusion criteria for "Concept" and "Context",1 did not meet the inclusion criteria for "Language", and 277 did not meet the updated exclusion criteria for "Source Type" following protocol deviation.

During screening, the full texts of previously selected sources were manually searched using the keyword "whey" (via Ctrl + F) to assess their relevance, which led to the discovery of four additional sources cited within those papers, which were not found during the primary search but met the inclusion criteria independently. These four sources were added to the review, resulting in a final set of 56 full-text papers for detailed analysis.

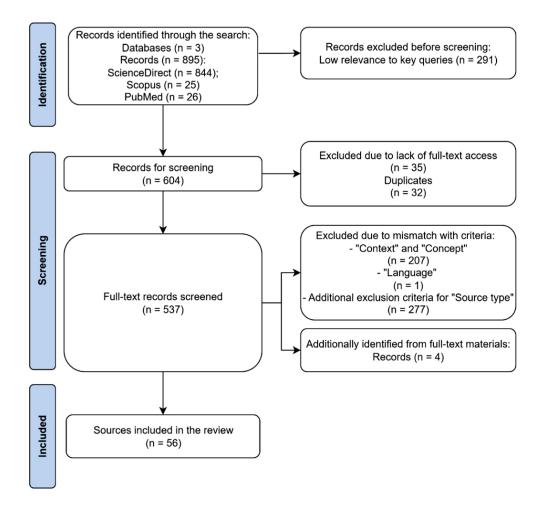
The PRISMA-ScR flow diagram illustrating the source selection process is presented in Figure 1.

An analysis of the excluded sources based on the "Concept" and "Context" criteria revealed that several studies didn't include practical examples, technological descriptions, or analyses of direct 3DP implementation, and only mentioned the potential of 3DP. In some cases, whey was mentioned solely for comparative purposes and was not the primary or one of the primary research objects. This significantly limited the value of such sources for analyzing whey-specific properties in the context of 3DP.

Despite the large number of publications on other materials used in 3DP ink formulations, no additional modifications were made to the initial search queries. This decision was based on preliminary analysis, which showed that whey-derived products are frequently used in combination with other food ingredients.

Figure 1

PRISMA-ScR Flow Diagram of the Source Selection Process for the Scoping Review



Visualization

All included sources were exported in RIS format and analyzed using the "VOSviewer" software, which allowed for the visualization of keyword co-occurrence and interrelations based on bibliographic data, providing a graphical representation of the scoping review structure. The minimum threshold for keyword occurrence was set at 2.

RESULTS

Description of Included Sources

As a result of the analysis, 16 keywords or keyword phrases were identified as occurring two or more times out of a total of 157 unique keywords. Their frequency of occurrence is illustrated in Figure 2.

Figure 2 indicates that the keywords from the included sources are consistent with the research questions of this review. These studies mainly focus on the rheological properties, texture, and microstructure, as well as the use of whey-derived components, such as whey protein isolate, in 3DP. The most commonly used materials for 3DP were emulsion gels, emulsions, and high internal phase emulsions (HIPEs).

The results of the chronological distribution analysis of the included sources are shown in Figure 3.

Although the review covered 15 years (2010–2025), included sources meeting the inclusion criteria were published from 2018 to 2025. The lack of relevant publications prior to 2018 is likely due to the fact that earlier research on 3DP fundamental technological aspects, with limited attention paid to the potential and prospects of utilizing

Figure 2 **Keyword Frequency Distribution**

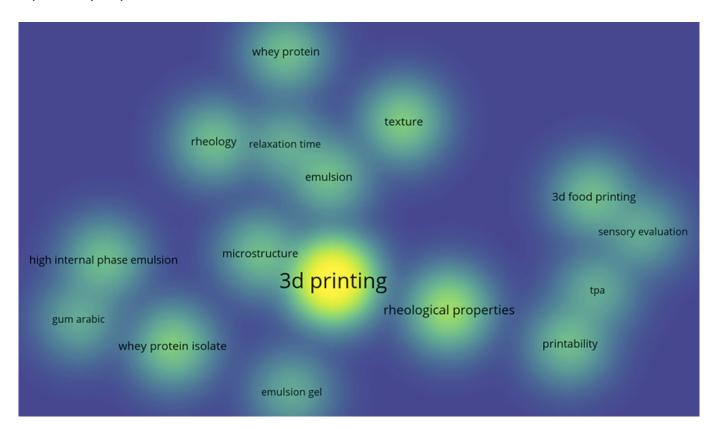
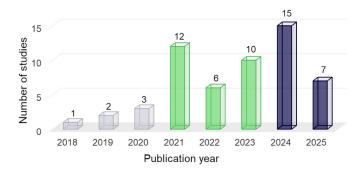


Figure 3

Chronological Distribution of Included Sources



secondary raw materials, such as whey, for the formulation of 3DP inks used in printed structures.

The highest number of relevant studies was published between 2021 and 2024, which can be due to the rapid development and increasing accessibility of additive manufacturing technologies in recent years. According to UnivDatos, the global 3D printing market was estimated at approximately USD 15.5 billion in 2023 and is expected to

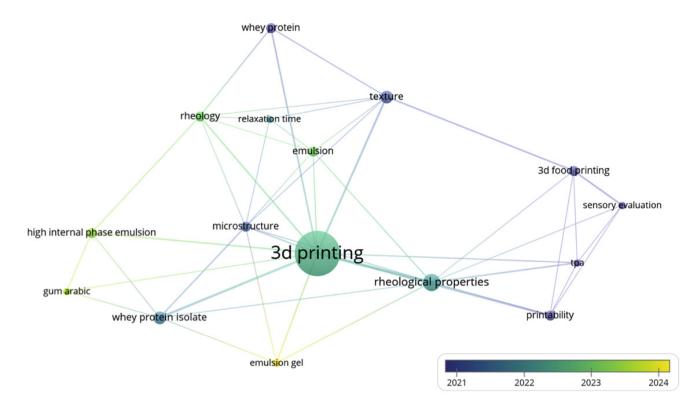
grow at a steady compound annual growth rate (CAGR) of about 19.5 % during the forecast period.

A keyword co-occurrence map with chronological overlay is presented in Figure 4.

Composition and Types of Ink Systems for 3DP

Analysis of the selected sources revealed that the primary objective in the context of using whey-derived products in 3DP is the development of materials with optimal printability, resulting in the creation of stable and practically applicable printed objects. While this overarching goal is shared across studies, it is pursued through different approaches: in some cases, by selecting and optimizing the composition and concentrations of ink formulations (Shi et al., 2025; Liu et al., 2018), in others, by applying various technological treatments—such as pH adjustment, heat treatment or mechanical processing—either before printing (Daffner et al., 2021; Uribe-Alvarez et al., 2023) or as post-processing of the printed structures (Llamas-Unzueta

Figure 4 **Keyword Co-occurrence Map with Chronological Overlay**



et al., 2022; Kamlow et al., 2021). These approaches may also be combined within a single study.

Nevertheless, the core task in developing a material for 3DP lies in the selection and justification of its formulation components. The physicochemical properties of these components determine the material's ability to form intermolecular interactions, which directly influence the struc-

tural characteristics of the final printed object (Daffner et al., 2021a). Accordingly, the first stage of analysis focused on reviewing the component composition of the ink formulations and identifying the types of systems developed, as presented in Table 2. The system type was determined based on the terminology used by the authors to describe their 3DP materials.

Table 2

Component Composition and System Type of Materials Used for 3D Printing

Other Main Ingredients	System Type	Reference
Whey protein isolate Micellar casein concentrate, cream (dairy fat) Suspensi		Daffner et al. (2021a)
Linseed oil, κ-carrageenan (0–0.8%)	Emulsion, emulsion gel	Shi et al. (2025)
α-Lactose monohydrate, deionized water	Not specified*	Fanghui Fan et al. (2022)
Micellar casein concentrate, deionized water	Suspension	Daffner et al. (2021b)
Milk protein concentrate, glycerol, xanthan gum	Paste	Liu et al. (2018)
Greek yogurt, gelatin, and sweetener	Gel	Riantiningtyas et al. (2021)
	Micellar casein concentrate, cream (dairy fat) Linseed oil, κ-carrageenan (0–0.8%) α-Lactose monohydrate, deionized water Micellar casein concentrate, deionized water Milk protein concentrate, glycerol, xanthan gum	Micellar casein concentrate, cream (dairy fat) Suspension Linseed oil, κ-carrageenan (0–0.8%) Emulsion, emulsion gel α-Lactose monohydrate, deionized water Not specified* Micellar casein concentrate, deionized water Suspension Milk protein concentrate, glycerol, xanthan gum Paste

Whey Component	Other Main Ingredients	System Type	Reference
	Soft cheese, obtained via heat-acid coagulation, maltitol	Composite semi-solid material	Bareen et al. (2021, 2023a, 2023b)
	Canola oil, corn starch, distilled water	Paste	Liu et al. (2021)
	Calcium chloride, water	Gel	Uribe-Alvarez et al. (2023)
	Sunflower oil, κ-carrageenan	Emulsion gel	Kamlow et al. (2021)
	Hydroxypropyl starch, carrageenan, corn oil, whole milk powder, cellulose, sodium alginate	Emulsion	Cai et al. (2022)
	Polyvinyl alcohol, glutaraldehyde solution, phosphate buffer, gentamicin sulfate, hydroxyapatite	Composite	Tut et al. (2022)
	Milk, soybean oil, non-emulsified liquid, crystalline vitamin D3 powder, sodium caseinate, and sodium azide	Nanoemulsion gel	Joshi et al. (2024)
	Potato starch, corn oil, curcumin, xanthan gum	Emulsion gel	Cheng et al. (2024)
	Gelatin, whipped cream, pasteurized eggs, sugar, citric acid	Gel	Chow et al. (2021)
	Rice starch, soybean oil, resveratrol, β-carotene	High internal phase emulsion, gel	Zheng et al. (2024)
	Gellan gum	Gel	Oliveira et al. (2020)
	Probiotic, epigallocatechin gallate, hydroxypropyl starch, resveratrol, whole milk powder, cellulose, sodium alginate, carrageenan, gelatin, sodium caseinate, etc.	Emulsion gel	Cai et al. (2023)
	к-carrageenan, sunflower oil, cinnamaldehyde, potassium chloride, water	Emulsion gel	Kamlow et al. (2022)
	Gum arabic, olive oil	High internal phase emulsion, gel	Kan et al. (2023)
	Soybean oil	Pickering high internal phase emulsion, gel	Liu et al. (2019)
	Soybean oil, transglutaminase, sodium chloride	Emulsion gel	Li et al. (2024)
	Asparagus fiber concentrate, oil	Emulsion gel	Lu et al. (2024)
	Soybean oil, fucoxanthin, glyceryl monostearate	Pickering high internal phase emulsion, gel	Shang et al. (2023)
	Potato starch, xanthan gum, curcumin	Gel	Shen et al. (2023)
	Corn starch	Gel	Xian et al. (2024)
	Gum arabic, sodium alginate	Hydrogel	Kan et al. (2024)
	Freeze-dried powder of Flammulina velutipes, olive oil, xanthan gum, etc.	Emulsion gel	Dong et al. (2024)
	Freeze-dried red cabbage powder, dried apple powder, sodium alginate, gum arabic	Gel	Ghazal et al. (2023)
	Micellar casein, soy protein isolate, pea protein isolate, titanium dioxide, graphite, chitosan, sodium caseinate, sunflower oil, etc.	Emulsion gel	W. Li et al. (2021)

Whey Component	Other Main Ingredients	System Type	Reference
	Micellar casein, microparticulated whey protein, whipped cream, sodium chloride, water	Emulsion gel	Sager et al. (2020)
	Guar gum, xanthan gum, gum arabic, medium-chain triglycerides	Emulsion gel	Feng et al. (2025)
	Riboflavin, xanthan gum	Paste	Araújo et al. (2025)
	к-carrageenan, konjac gum	Gel	Kong et al. (2025)
	β-carotene, guar gum, locust bean gum, xanthan gum, gum arabic	Emulsion gel	Li et al. (2023)
	Gelatin, alginate	Hydrogel	Sümbelli et al. (2021)
	Lotus root	Composite gel	Wang et al. (2025)
	Soybean oil, proanthocyanins, curcumin	High internal phase Pickering emulsions	Ji et al. (2025)
	Antarctic krill oil, carboxymethyl chitosan	Emulsion-templated oleogels	Zhao et al. (2025)
	Methacrylic anhydride	Hydrogels	Hu et al. (2022)
	Peach gum polysaccharide complex, telechelic DNA	Emulsion gel	R. Zhang et al. (2024)
Fibrillar whey protein isolate	Fructooligosaccharides, gellan gum, tributyrin, probiotic	Composite hydrogel	Zhang et al. (2024)
isolate	Soybean oil, starches, curcumin	Composite emulsion gel	Z. Wang et al. (2024)
	Zein, corn oil, lycopene	Pickering high internal phase emulsion	Xia et al. (2024)
Whey protein concentrate	Konjac flour, curdlan, sodium bicarbonate	Gel	Du et al. (2021)
concentrate	Sunflower oil, beeswax, and potato starch	Oleogel	Shi et al. (2021)
	Pumpkin flour, dried spinach, sorghum flour, dried purple potato, xanthan gum	Paste	Zheng et al. (2021)
	Calcium caseinate, high-fructose corn syrup, oil (medium-chain triglycerides), glycerol, chocolate	Paste	Zhu et al. (2021)
	Biodegradable poly lactic acid (PLA)	Solution	Kayadurmus et al. (2024)
Whey powder	Water	Paste	Llamas-Unzueta et al. (2022, 2024)
β-Lactoglobulin	Gelatin, sodium alginate, dopamine hydrochloride, etc.	Gel	Ghorbani et al. (2024)
Lactoferrin	Pectin, κ -carrageenan, curcumin, sunflower oil, β -carotene, oil (medium-chain triglycerides), oat β -glucan, chitosan, epigallocatechin gallate	High internal phase emulsion, hydrogels	Xu et al. (2023)
	Polycaprolactone, WPI	Intermixed powder blend biomaterial	Hewitt et al. (2019)

Note. * Not specified — no information provided in the source ** High internal phase emulsion — emulsion system with a high volume fraction of internal phase

The type of systems used for extrusion-based 3DP is limited by the fact that inks must have specific rheological properties to pass through the nozzle and ensure a balance between the flowability of the material and its ability to retain its shape after extrusion (Hussain et al., 2021). This means that the material should exhibit shear-thinning behavior (becoming less viscous when stress is applied) along with sufficient structural stability after layer deposition. A literature review revealed a variety of systems used in 3DP, including suspensions, emulsion gels, hydrogels, pastes, and composite systems. At the stage of ink formulation development, the material used was mainly in the form of emulsions or suspensions (Daffner et al., 2021a; Shi et al., 2025), after which technological operations facilitated the transition to a more structured state (mainly a gel), providing the necessary viscosity and mechanical strength for printing.

Regardless of system type, literature overview shows that in 76% of the reviewed studies, whey protein isolate (WPI) was used as the whey-derived component; 5% used WPI in its fibrillar form, 9% used whey protein concentrates (WPC), 4% applied whey powder, 2% and 4% used individual whey proteins such as β -lactoglobulin and lactoferrin, respectively. The prevalence of WPI in formulations may be due to a number of its advantages, including high nutritional value, emulsifying properties, digestibility and bioavailability, biocompatibility, and gelation potential (Sager et al., 2020; Tut et al., 2022; Cai et al., 2023; Riantiningtyas et al., 2024; Shi et al., 2025). In the studies by Daffner et al. (2021b) and Bareen et al. (2021), the addition of WPI into casein-containing matrices (milk protein concentrate, skim milk) resulted in gel stabilization through disulfide bond formation, which improved the strength of printed constructs. A similar effect was found by Dong et al. (2024), who compared WPI with plant protein concentrates that cannot form disulfide bonds.

Conversely, studies by Riantiningtyas et al. (2024) and Liu et al. (2018) used WPI to reduce viscosity and weaken the gel structure, which may appear disadvantageous, but in fact it facilitates extrusion through narrow nozzles of 3D printers. These examples highlight the unique properties of WPI, as it can play different roles depending on the composition of the formulation. Thus, WPI served as an emulsifier in studies by Liu et al. (2021), Kamlow et al. (2021, 2022), Cai et al. (2022), Joshi et al. (2024), Zheng et al. (2024), and Lu et al. (2024); as a stabilizer in works by Oliveira et al. (2020), Liu et al. (2019), and Shang et al. (2023).

However, despite these advantages, several research groups also reported adverse effects associated with the addition of WPI or increasing its concentration in formulations, namely limited stability (Sager et al., 2020), reduced elasticity (Liu et al., 2018), impaired textural properties (Liu et al., 2021), inability to fill interdisperse voids (Li et al., 2021), low water-holding capacity (Dong et al., 2024), and poor shape retention of complex architectures (Xian et al., 2024).

To overcome these limitations, researchers have explored various strategies to modulate WPI properties and develop competitive 3DP inks. The structure and stability of WPI gels can be controlled by additives, such as polysaccharides (xanthan gum, guar gum, gellan gum, gum arabic, carrageenan, etc.), which are widely used in this research field (Liu et al., 2018; Kamlow et al., 2022; Shen et al., 2023; Cheng et al., 2024). The Maillard reaction between whey proteins and polysaccharides is one of the mechanisms of gel stabilization according to Kan et al. (2023). Moreover, Li et al. (2024) demonstrated that the addition of transglutaminase also can be a way to improve gels extrudability. Transglutaminase improved gel structure ordering and optimized water distribution within the gel network. According to the authors, transglutaminase markedly increased 3DP precision.

Other formulation strategies to improve gel properties included the addition of dairy fat fractions (Sager et al., 2020; Daffner et al., 2021a), the use of fibrillar WPI (Xia et al., 2024; Z. Wang et al., 2024), and the inclusion of microparticulated whey proteins as inert fillers that do not participate in bonding (Sager et al., 2020).

Besides formulation-based approaches, other printing technologies can produce materials with different structural and functional features compared to extrusion. For example, Hewitt et al. (2019) used the melt electrowriting (MEW) printing technique, which helped create precise pore shapes and higher porosity in the final materials. These materials also showed improved bioactivity.

Technological operations (pH adjustment, heat treatment, ultrasonication, and high-pressure homogenization) were also used to modify the properties of 3DP materials and printed objects (Daffner et al., 2021a, 2021b; Du et al., 2021; W. Li et al., 2021; Z. Wang et al., 2024; Shi et al., 2025). In protein-based hydrogels, the in situ cross-linking method called Amino Acid (monomer) Decorated and Light Underpinning Conjugation Approach (ANADOLUCA) led to

the production of non-cytotoxic whey protein gels with improved stability while preserving cell viability (Sümbelli et al., 2020). Heat treatment and pH adjustment were used to control the state of proteins, namely their denaturation, interactions with other components, and the formation of complexes. Mechanical processing was primarily aimed to obtain homogeneous mixtures and prevent phase separation, reduce fat particle size, increase surface area, and facilitate the formation of emulsions. As post-processing steps, Llamas-Unzueta et al. (2022, 2024) used carbonisation and various drying techniques: microwave, infrared, and hot air drying (Shen et al., 2023) to stabilize printed

structures or to modify sensory characteristics (Ghazal et al., 2023). Despite differences in enhancement strategies, most studies evaluated a similar set of key material properties using standardized methodological approaches.

Properties of 3DP Materials and Analytical Methods

Each material property assessed in the reviewed studies addresses a specific requirement for 3DP suitability and may be investigated using several analytical techniques (Table 3).

Table 3

Analyzed properties and methods used for characterization of 3DP inks and printed constructs

Indicators or properties under study	Method and equipment	Reference
Rheology		
Storage modulus (G'), loss modulus (G"), yield stress, and/or other rheological tests.	Rheometry, Kinexus Pro, DHR, HAAKE, Discovery HR-3, MCR, etc.	Daffner et al. (2021a), Daffner et al. (2021b), Du et al. (2021), Z. Wang et al. (2024), Shi et al. (2025), Fan et al. (2022), Liu et al. (2018), Riantiningtyas et al. (2021), Shi et al. (2023a), Bareen et al. (2024), Bareen et al. (2021), Uribe-Alvarez et al. (2023), Kamlow et al. (2021), Joshi et al. (2024), Llamas-Unzueta et al. (2022), Cheng et al. (2024), Chow et al., (2021), Zheng et al. (2024), Oliveira et al. (2020), Zheng et al. (2021), Liu et al. (2019), Kan et al. (2023), Li et al. (2024), Lu et al. (2024), Shang et al. (2023), Shen et al. (2023), Xian et al. (2024), Dong et al. (2024), Ghazal et al. (2023), Li et al. (2025), Li et al. (2023), Sümbelli et al. (2021), Wang et al. (2025), Li et al. (2025), Zhao et al. (2025), Hu et al. (2022), R. Zhang et al. (2024)
Sol–gel transition temperature (T _{sol-gel})*	Rheometry, Kinexus Pro	Daffner et al. (2021a); Daffner et al. (2021b)
Viscosity	Rheometry, Kinexus Pro, DHR, HAAKE, Discovery HR-3, MCR, etc.	Shi et al. (2025), Fan et al. (2022), Liu et al. (2018), Bareen et al. (2021), Bareen et al. (2023a), Liu et al. (2021), Bareen et al. (2023b), Llamas-Unzueta et al. (2022), Chow et al., (2021), Oliveira et al. (2020), Liu et al. (2019), Kan et al. (2023), Shang et al. (2023), Shen et al. (2023), Dong et al. (2024), Ghazal et al. (2023), Xu et al. (2023), R. Zhang et al. (2024)
Thixotropy	Rheometry, DHR, HAAKE, MCR	Shi et al. (2025), Zheng et al. (2024), Liu et al. (2019), Shang et al. (2023), Xian et al. (2024)
Pseudoplasticity (flow curves)	Rheometry, MCR	Joshi et al. (2024)
Creep-recovery behavior	Rheometry, DHR, MCR	Riantiningtyas et al. (2021), Bareen et al. (2023a), Zheng et al. (2024), Sager et al. (2020)
Recovery index	Rheometry, MCR	Bareen et al. (2021)
Relaxation time	Rheometry, HAAKE	Fan et al. (2022)
	Low-field nuclear magnetic resonance (low-field NMR) MesoMR23–060H	Zhang et al. (2024), Shang et al. (2023)
Extrudability	Measurement of mechanical force using a universal testing machine (Zwick Roell Z010) equipped with a 50 N load cell	Uribe-Alvarez et al. (2023)

Indicators or properties under study	Method and equipment	Reference
Texture Profile		
Textural properties (gel strength, hardness, crispness, adhesiveness, elasticity, etc.)	Texture analysis, TA-XT, TA-XTC, Texture Pro, etc.	Shi et al. (2025), Fan et al. (2022), Liu et al. (2018), Z. Wang et al. (2024), Riantiningtyas et al. (2021), Shi et al. (2021), Bareen et al. (2021), Bareen et al. (2023), Liu et al. (2021), Uribe-Alvarez et al. (2023), Kamlow et al. (2021), Cheng et al. (2024), Chow et al. (2021), Zhu et al. (2021), Kamlow et al. (2022), Liu et al. (2019), Zhang et al. (2024), Kan et al. (2022), Li et al. (2024), Xian et al. (2024), Kan et al. (2023), Sager et al. (2020), Feng et al. (2025), Kong et al. (2025), Li et al. (2023), Wang et al. (2025), Zhao et al. (2025)
Particle Characteristics and Interactions		
Zeta (ζ) potential and/or particle size (including droplet size), polydispersity index	Laser diffraction method, Malvern Mastersizer, Zetasizer SYNC, etc.	Daffner et al. (2021a), Daffner et al. (2021b), Bareen et al. (2023b), Joshi et al. (2024), Zheng et al. (2024), Shi et al. (2025), Cai et al. (2023), Kamlow et al. (2022), Liu et al. (2019), Zhang et al. (2024), Kan et al. (2023), Li et al. (2024), Shang et al. (2023), Kan et al. (2024), Ghazal et al. (2023), Xu et al. (2023), W. Li et al. (2021), Feng et al. (2025), Araújo et al. (2025), Ji et al. (2025), R. Zhang et al. (2024)
	Nuclear Magnetic Resonance (NMR) Spectroscopy	Kamlow et al. (2022)
	Droplet images (bright-	Zhao et al. (2025)
	field microscopy) were analyzed using Nano Measurer software	
Analysis of Components Interactions	Fourier-transform infrared (FTIR) spectroscopy, Nicolet iS50, Jasco, etc.	Shi et al. (2025), Tut et al. (2022), Cheng et al. (2024), Ghorbani et al. (2024), Zheng et al. (2024), Li et al. (2024), Shang et al. (2023), Shen et al. (2023), Kan et al. (2024), Dong et al. (2024), Feng et al. (2025), Kayadurmus et al. (2024), Wang et al. (2025), Hu et al. (2022), R. Zhang et al. (2024)
	Circular Dichroism Spectroscopy	Shang et al. (2023), Hu et al. (2022)
	Nuclear Magnetic Resonance Spectroscopy	Hu et al. (2022)
	Surface hydrophobicity: bromophenol blue (BPB) binding method at 25 °C; absorbance at 596 nm)	Wang et al. (2025)
	Intermolecular force test: protein solubility in selective denaturants (NaCl, urea, β-mercaptoethanol) to determine contributions of ionic, hydrogen, hydrophobic, and disulfide interactions	Wang et al. (2025)
Interfacial Properties (oil-water interfacial tension, gas-liquid surface tension, etc.)	Contact angle goniometer and oscillating drop device	Shi et al. (2025), Zheng et al. (2024), Shang et al. (2023)
	DSA-30 Drop Shape Analyzer for 3-phase contact angle; interfacial rheometer (drop method) and DCAT21 with Wilhelmy plate method for gas–liquid surface tension and oil–water interfacial tension.	Ji et al. (2025)
	Protein adsorption rate (AP) and surface load (Γ) at oil–water interface determined by BCA assay after centrifugation and filtration	Ji et al. (2025)

Indicators or properties under study	Method and equipment	Reference
Microstructure	Confocal laser scanning microscopy, such	Daffner et al. (2021a),
	as Leica, Olympus, and Zeiss.	Shi et al. (2025), Liu et al. (2018), Xia et al. (2024), Bareen et al (2023a), Kamlow et al. (2021), Chow et al., (2021), Zheng et al. (2024), Oliveira et al. (2020), Zhang et al. (2024), Kan et al. (2023), Li et al. (2024), Lu et al. (2024), Shang et al. (2023), Kar et al. (2024), Feng et al. (2025)
	Cryo-transmission electron microscopy, Tecnai G2 F30 (200 kV) with CETA CMOS detector	Daffner et al. (2021a)
	Transmission electron microscopy (TEM)	Zhang et al. (2024), Kan et al. (2024)
	Cryo-scanning electron microscopy (Cryo- SEM), Hitachi, Magellan, etc.	Shi et al. (2025), Cheng et al. (2024), Zheng et al. (2024), Li et al. (2024), Lu et al. (2024)
	Scanning electron microscopy (including field-emission SEM), Phenom Pro, Zeiss, etc.	Z. Wang et al. (2024), Bareen et al. (2021), Tut et al. (2022), Bareen et al. (2023b), Joshi et al. (2024), Llamas-Unzueta et al. (2022), Ghorbani et al. (2024), Liu et al. (2019), Shang et al. (2023), Kan et al. (2024), Dong et al. (2024), Ghazal et al. (2023), Hewitt et al. (2019), Kayadurmus et al. (2024), Sümbelli et al. (2021), Wang et al. (2025), Ji et al. (2025), Hu et al. (2022)
	Fluorescence microscopy, Axio Vert A1, etc.	Du et al. (2021), Shi et al. (2021), Liu et al. (2019), Shang et al. (2023), Ghazal et al. (2023), Hewitt et al. (2019)6 Ji et al. (2025), R. Zhang et al. (2024)
	Optical microscopy, such as Leica and Olympus, etc.	Xia et al. (2024), Kan et al. (2023), Xian et al. (2024), Kan et al. (2024), W. Li et al. (2021), Ji et al. (2025)
	Bright-	Zhao et al. (2025)
	field microscopy	
	X-ray diffraction (XRD)	Tut et al. (2022), Joshi et al. (2024), Dong et al. (2024), Wang et al. (2025)
	Polarized light microscopy	Cai et al. (2023)
	Laser confocal Raman spectroscopy	Wang et al. (2025)
Protein Composition	Sodium dodecyl sulfate–polyacrylamide gel electrophoresis (SDS-PAGE)	Daffner et al. (2021a), Daffner et al. (2021b), Kan et al. (2024)
Water Holding Capacity	Centrifugation and calculation based on the formula	Shi et al. (2025), Z. Wang et al. (2024), Dong et al. (2024), Kong et al. (2025), Li et al. (2023)
Oil Holding Capacity	Centrifugation and calculation based on the formula	Zheng et al. (2024), Zhao et al. (2025)
Water Distribution	Magnetic resonance imaging (MRI), MesoMR23–060H-I	Shi et al. (2025)
	Low-field nuclear magnetic resonance (LF-NMR), MicroMR20–030V-I, PQ001, etc.	Du et al. (2021), Liu et al. (2018), Zheng et al. (2021), Li et al. (2024), Shen et al. (2023), Dong et al. (2024), Kong et al. (2025), Li et al. (2023), Wang et al. (2025)
Thermophysical Properties (Glass transition, melting, crystallization,	Differential scanning calorimetry (DSC)	Fan et al. (2022), Shi et al. (2021), Kamlow et al. (2021), Tut et al. (2022), Zheng et al. (2024), Li et al. (2024), Liu et al. (2019)
gelation temperatures)	Thermogravimetric analysis (TGA) from 20 to 600 °C at 10 °C/min under nitrogen; TG and DTG curves recorded.	Wang et al. (2025)

Indicators or properties under study	Method and equipment	Reference
Porosity, or/and Pore Size, or/and strut size	Nitrogen adsorption/desorption at low temperatures, BELSORP-MR6	Fan et al. (2022)
	SEM images analysis with or without using ImageJ (ND plugin) and DiameterJ.	Hewitt et al. (2019). Kayadurmus et al. (2024)
	Mercury intrusion porosimetry	Llamas-Unzueta et al. (2022)
Stability of 3DP Materials and Printed Obje	ects	
Storage Stability	Storage at 4 °C for 90 days; visual inspection	Shi et al. (2025)
	Storage at 4 °C for 30 days; visual inspection	Xia et al. (2024)
	Storage at 25 °C for 14 days	Shang et al. (2023)
	Storage at 25 °C for 1 year	Xu et al. (2023)
	Storage at 4 °C for 7 days, photographing	Ji et al. (2025)
	Swelling and/or degradation testing with or without calculation	Tut et al. (2022), Ghorbani et al. (2024), Feng et al. (2025), Hewitt et al. (2019), Kayadurmus et al. (2024), Sümbelli et al (2021)
	Biodegradation analysis with Qubit® Protein Assay Kit; protein release monitored over 21 days and expressed as % of loaded protein	Hewitt et al. (2019)
Liquid Properties in Materials	Syneresis test, measurements, and calculations	Kamlow et al. (2022)
	Water loss test, measurements, and calculations	Xian et al. (2024)
Stability Evaluation	Assessment of the effect of salt concentration on gel stability and microstructure	Shang et al. (2023)
Oxidation Stability	Peroxide value (POV) determination for primary lipid oxidation products and TBARS assay to determine secondary lipid oxidation products (malondialdehyde equivalents)	Ji et al. (2025), Zhao et al. (2025)
Heating Stability/ Pasteurization properties	Heating at 90°C in a water bath for 30 minutes, followed by visual inspection and temperature scanning using a HAAKE MARS60, Thermo Fisher.	Shi et al. (2025)
	Heating at 75 °C for 30 minutes, followed by cooling to 25 °C in an ice bath, and visual inspection	Xia et al. (2024)
	Heating from room temperature to 800 °C at 20 °C/min under nitrogen using TGA550 (TA Instruments, USA)	Feng et al. (2025)
	Heating from 50 °C to 95 °C at 12 °C/min, holding 2.5 min, then cooling to 50 °C at the same rate using RVA-Super analyzer; viscosity parameters (PV, TV, BV, FV, SV, PT, Ptemp) recorded.	Wang et al. (2025)

Indicators or properties under study	Method and equipment	Reference
Freeze–Thaw Stability and Thawing Behavior	Freezing at –20 °C for 24 h, thawing at 25 °C for 2 h; visual inspection	Shi et al. (2025)
	Freezing at -20 °C for 18 h, thawing at 20 °C for 6 h; visual inspection	Xia et al. (2024)
	Freezing at -20 °C for 4 h, thawing at 25 °C for 6 h; microstructure and relaxation time analysis	Shang et al. (2023)
	Differential scanning calorimetry, Pyris Diamond	Liu et al. (2019)
Centrifugation Stability	Visual and calculated evaluation after centrifugation using masses before and after	Xia et al. (2024), Lu et al. (2024), Ji et al. (2025)
Stiffness (Storage Modulus E')	Dynamic mechanical analyzer, Mettler-Toledo	Fan et al. (2022)
Resistance to Mechanical Load (Loss Modulus E")	Dynamic mechanical analyzer, Mettler-Toledo	Fan et al. (2022)
Printability and/or Geometric and Physical Characteristics of printed objects	Visual evaluation, photographic analysis, and/or quantitative measurements	Z. Wang et al. (2024), Uribe-Alvarez et al. (2023), Riantiningtyas et al. (2021), Bareen et al. (2023b), Joshi et al. (2024), Chow et al., (2021), Zheng et al. (2021), Lu et al. (2024), Araújo et al. (2025), Kong et al. (2025), Li et al. (2023), Wang et al. (2025)
Turbiscan Stability Index (TSI)	Turbiscan analysis and TSI calculation	Feng et al. (2025), Zhao et al. (2025), R. Zhang et al. (2024)
Other Specific Methods		
Mechanical Properties (Strength, Compressibility)	Compression testing, SHIMADZU EZ-LX, Instron Model 8562 etc.	Tut et al. (2022), Llamas-Unzueta et al. (2022), Kayadurmus et al. (2024), Sümbelli et al. (2021), Hu et al. (2022)
	Torque measurements	Llamas-Unzueta et al. (2024)
Encapsulation Efficiency/Drug Release Efficiency and/or Substance Stability	Spectrophotometry and calculations, Lambda 35 UV-VIS, TU-1810PC, etc.	Tut et al. (2022), Joshi et al. (2024), Cheng et al. (2024), Zheng et al. (2024), Shang et al. (2023), Shen et al. (2023), Araújo et al. (2025), Kayadurmus et al. (2024), Li et al. (2023), Ji et al. (2025), Ji et al. (2025)
	Plate count method for probiotics, HPLC for epigallocatechin gallate	Cai et al. (2023)
Bioactive Compound Release	An in vitro digestion system equipped with an automatic titrator, incubator, and other accessories, with or without calculations	Cai et al. (2023), Shang et al. (2023), Feng et al. (2025)
	In vitro digestion, centrifugation, and fluorescence spectroscopy to determine Rb bioaccessibility (%)	Araújo et al. (2025)
	Centrifugation of intestinal digesta to isolate micellar phase, extraction with methanol/MTBE, and AST quantification to calculate bioaccessibility (%)	Zhao et al. (2025)
Digestive characteristics	In vitro digestion with or without calculation of the degree of hydrolysis	Feng et al. (2025), Araújo et al. (2025), Zhao et al. (2025)

Continuation of the Table 3

Indicators or properties under study	Method and equipment	Reference
Cell Studies (Adhesion, Viability, and	Serial dilution plate count method	Cai et al. (2023)
Morphology)	Colorimetric MTT assay and\or scanning electron microscopy	Tut et al. (2022), Hewitt et al. (2019), Araújo et al. (2025)
	Live/Dead assay (Calcein AM, EthD-1) and fluorescence microscopy to assess cell viability (days 1, 3, 7).	Kayadurmus et al. (2024)
	Cell proliferation assay using PrestoBlue, ALP activity analysis	Ghorbani et al. (2024)
Cytotoxicity	MTT assay	Kayadurmus et al. (2024)
	CCK-8 assay	Hu et al. (2022)
Wound Healing Capacity	In vitro wound healing assay (scratch test); HaCaT, Nhdf, and co-culture; wound width measured at 0 and 24 h using CellSens software (Leica DM IL)	Hewitt et al. (2019)
Elemental Analysis	LECO analyzer	Llamas-Unzueta et al. (2022), Llamas-Unzueta et al. (2024)
Mineral Content in Ash	Elemental analysis via X-ray fluorescence spectroscopy, Bruker	Llamas-Unzueta et al. (2022)
Protein Solubility	Protein solubility in solutions disrupting specific interactions (electrostatic, hydrogen, hydrophobic, disulfide) to determine their contribution to the sample's structure.	Li et al. (2023)
Color	Color meter (CM-700d)	Li et al. (2023)
Volatile organic compounds	GC-IMS	Zhao et al. (2025)
In vivo tests	Rat Subcutaneous Implantation Test and Rat Myocardial Infarction Model with Hydrogel Injection	Hu et al. (2022)

Assessment of rheological and textural properties, microstructure, and particle size measurements were the most commonly applied analytical methods in the reviewed studies, highlighting their importance in determining material suitability for 3DP. Specifically, 77% of the studies evaluated the yield stress and/or the storage (G') and loss (G") moduli. The storage modulus is a parameter that reflects the ability of a material to accumulate energy during deformation (Riantiningtyas et al., 2021), and it provides understanding of a material's ability to retain shape after printing (Daffner et al., 2021a, 2021b; Zheng et al., 2021).

In turn, Zheng et al. (2021) used texture profile analysis to calculate Young's modulus (the slope of the stress–strain curve), which, according to the authors, turned out to be a

key indicator of printability. Similar findings were reported by Sager et al. (2020) and Kan et al. (2024).

Microstructural analysis, conducted using various microscopy techniques and X-ray diffraction in 69% of the reviewed publications, aimed to evaluate component interactions within the 3DP matrix, assess the degree of structural order, and examine the development of the gel network. Additionally, 38% of the studies included zeta potential and particle size analysis. These parameters allowed to evaluate the electrostatic stability of the material and particle size distribution, which is crucial to predict the material's colloidal stability, flow behavior, and ability to retain its shape after extrusion.

Taken together, a basic integrated approach to evaluating 3DP material suitability should comprise rheological, textural, and microstructural assessments — analytical procedures that are generally cost-effective and widely accessible. In addition, it was found that some researchers (Xia et al., 2024; Shang et al., 2023) proposed specific ways and conditions to evaluate the stability and robustness of printed constructs, including freeze-thaw cycles, photographic or visual monitoring, and measurement of geometrical or physical parameters after storage. Also, other unpopular ways to evaluate the stability of final products or objects were found. They included an evaluation of oxidation stability for gels with oils (Ji et al., 2025; Zhao et al., 2025), heating stability (Xia et al., 2024; Shi et al., 2025; Feng et al., 2025; Wang et al., 2025), and centrifugation stability (Xia et al., 2024; Lu et al., 2024; Ji et al., 2025).

In the case of developments with specific potential applications, namely for the chemical industry (Llamas-Unzueta et al., 2024), delivery of bioactive compounds in functional foods (Joshi et al., 2024; Cheng et al., 2024), or tissue engineering (Tut et al., 2022; Ghorbani et al., 2024), specific methods were used (analysis of elemental composition, release efficiency of bioactive compounds, and cell-material interactions). Additionally, other advancements at the junction of medicine and food science used uncommon (for food products) methods like cytotoxicity assessment with MTT and CCK-8 assays, which help determine the biocompatibility and safety of the materials obtained (Kayadurmus et al., 2024; Hu et al., 2022). In vivo testing on rats to evaluate

the biological response in a physiological environment after injections of the developed hydrogel for 3D printing was also performed in the study by Hu et al. (2022).

Application Areas of Whey Protein-Based 3DP Inks

Due to their versatility and multifunctionality, whey proteins have been applied across six key areas of 3D printing, spanning both the food industry and biomedical fields (Table 4).

The category of "personalized or customized products" combines two main tasks: the development of nutrition tailored to the individual nutritional needs of consumers (Kamlow et al., 2021; Kan et al., 2023), and taking into account their aesthetic expectations regarding the shape, appearance, and texture of the product, as discussed by Liu et al. (2019) and Chow et al. (2021). In some cases, personalization integrates both objectives into a single product (Zhang et al., 2024). According to the authors, functional and personalized products often aim to achieve common nutritional goals, including addressing micronutrient deficiencies, modulating physiological functions through the regular intake of bioactive-enriched foods, and reducing the content of specific components, such as fat, sugar, or salt. Some studies focus on creating products for specific consumer groups, for instance, for people with dysphagia. Z. Wang et al. (2024) conducted a special test using the

Table 4

Application areas of whey protein-based 3DP inks

Application Area	Examples of Studies
Personalized or Customized Products	Du et al. (2021), Daffner et al. (2021a), Daffner et al. (2021b) Shi et al. (2025), Riantiningtyas et al. (2024), Liu et al. (2018), Liu et al. (2021), Bareen et al. (2023a), Kamlow et al. (2021), Joshi et al. (2024), Chow et al. (2021), Zheng et al. (2024), Kamlow et al. (2022), Liu et al. (2019), Zhang et al. (2024), Kan et al. (2023), Lu et al. (2024), Kong et al. (2025
Functional Food Products	Fan et al. (2022), Joshi et al. (2024), Cheng et al. (2024), Cai et al. (2023), Kan et al. (2023), Shang et al. (2023), Kan et al. (2024), Feng et al. (2025), Araújo et al. (2025), Li et al. (2023), Ji et al. (2025)
Food for Individuals with Dysphagia	Z. Wang et al. (2024), Zhang et al. (2024), Kan et al. (2023), Li et al. (2024), Wang et al. (2025)
Printable Foods or Edible Materials	Bareen et al. (2021), Uribe-Alvarez et al. (2023), Cai et al. (2022), Chow et al. (2021), Oliveira et al. (2020), Zhu et al. (2021), Shang et al. (2023), Xian et al. (2024), Ghazal et al. (2023), Xu et al. (2023), Sager et al. (2020), W. Li et al. (2021), Liu et al. (2018), Shi et al. (2021), Dong et al. (2024)
Tissue Engineering, Biomedicine, and Pharmaceutics	Tut et al. (2022), Llamas-Unzueta et al. (2022), Ghorbani et al. (2024), Oliveira et al. (2020), Xu et al. (2023), W. Li et al. (2021), Hewitt et al. (2019), Kayadurmus et al. (2024), Hu et al. (2022)
Chemical Industry	Llamas-Unzueta et al. (2024)

IDDSI (International Dysphagia Diet Standardization Initiative) method to assess the suitability of the developed printed products as nutrition for people with dysphagia. The same was done by Kan et al. (2023), Zhang et al. (2024), and Li et al. (2024).

In the production of functional and personalized foods, whey proteins can serve as key components for the delivery and protection of bioactive compounds. The reason is they are able to ensure stabilization, release control, and improved bioavailability of bioactive substances. The study by Zhang et al. (2024) demonstrated the protective properties of WPI. WPI in the formulation of the developed 3DP foods prevented inactivation of probiotics during printing and storage. It corresponds with results of Joshi et al. (2024) who developed WPI-based nanoemulsion gels that encapsulated vitamin D3, maintaining structural stability during both storage and extrusion through the printer nozzle. Xia et al. (2024;), Zheng et al. (2021), Shen et al. (2023) did not indicate the intended application of their developed 3DP inks. However, based on their formulations, main objectives, and results, these gels could potentially serve as delivery systems or as matrices for 3D-printed food products.

In addition to food applications, 3DP technologies based on whey proteins have found promising uses in biomedicine, tissue engineering, and industrial applications. For example, whey proteins have been used in the creation of scaffold structures intended for bone tissue regeneration. Research by Tut et al. (2022) showed that WPI improves biocompatibility and supports osteoblast differentiation, which is critical for effective bone formation. In turn, Ghorbani et al. (2024) used β -lactoglobulin (β -LG) as a coating for polymer scaffolds, which improved their stability and bioactivity. Scaffolds incorporating WPI and β -LG also demonstrated the ability to release therapeutic agents in a controlled manner, suggesting their potential for treating bone defects. In other studies at the intersection of medicine and food science, melt-electrowritten PCL scaffolds were created for deep tissue dermal regeneration (Hewitt et al., 2019); scaffolds with WPC and PLA that have increased cell survival, high swelling, and degradation rates for wound healing applications (Kayadurmus et al., 2024); hydrogels cross-linked through a non-cytotoxic method with high biocompatibility for patient-specific tissue constructs (Sümbelli et al., 2020); and methacrylated whey protein hydrogels with good in vivo compatibility and therapeutic benefits in a myocardial infarction model (Hu et al., 2022).

Additional applications of whey protein-based 3DP structures include the development of porous carbon materials suitable for biomedical use. Llamas-Unzueta et al. (2022) introduced a carbonization technique for whey-based pastes to produce highly porous carbon scaffolds, demonstrating promise for tissue engineering applications. In a further study by Llamas-Unzueta et al. (2024), this method was adapted to create catalytic stirrers with permeable blades. These structures can be used in catalysis, water and gas purification processes, and in the development of novel biocompatible materials.

Hence, research on whey protein-based 3DP systems extends far beyond food technology and offers exciting opportunities in biomedicine, tissue engineering, and advanced manufacturing.

DISCUSSION

The conducted analysis is embedded in the broader global effort to find sustainable solutions for the utilization of whey, a by-product of dairy manufacturing whose volumes are steadily increasing in parallel with industry growth. In this context, 3DP is not only a key Industry 4.0 technology but also a tool for adding value to secondary raw materials, aligning with current ecological and economic trends.

The main findings of this review confirm the widespread use of whey proteins, particularly whey protein isolate (WPI), in 3DP formulations, attributed to their numerous advantages, including bioavailability, high nutritional value, biocompatibility, gelling capability, and capacity for hierarchical molecular assembly. The ability of WPI to form stable gels, regulate viscosity, and build structural scaffolds makes it an auspicious material for 3DP inks. However, the number of studies on WPI, with a relatively limited focus on other whey-derived components (e.g., WPC, dried whey, β-lactoglobulin, lactoferrin), highlights a gap in the exploration of alternative whey protein sources. Hydrolysates, for instance, with their bioactivity and modifiable functional properties, may expand the application spectrum of 3DP materials by improving solubility or introducing specific benefits.

The findings on material property analysis largely align with results from other recent reviews (N. Li et al., 2021; Y. Liu et al., 2024; Feng et al., 2024; Wu et al., 2024), which also identified rheology, texture profile, microstructure, and molecular interactions as key analytical blocks, while

emphasizing the importance of assessing final printed constructs. However, unlike previous reviews, the present study provides a detailed examination of the specific methods and equipment used in empirical studies, thereby offering practical guidance for research teams planning experimental workflows.

Among the two primary research avenues identified, food applications (personalized and functional nutrition) dominate, likely due to their stronger foundation within food science and the accessibility of analytical methods. The second prominent trend is the growing interest in biomedical applications such as scaffold systems for tissue engineering, where whey proteins serve as a bridge between the food and healthcare sectors. Nonetheless, successful integration of these technologies requires deeper exploration of methods for modulating the properties of whey-based 3DP materials throughout different processing stages, as well as interdisciplinary collaboration.

Review limitations

This review discusses only selected approaches to controlling the properties of whey-based 3D printing (3DP) materials reported in empirical studies. Individual whey proteins were not included in the search queries because the word "whey," which was included in the search queries, is usually mentioned in the description of these proteins. Also, it does not cover sources with limited information on whey in the context of 3DP and on individual proteins, although preliminary screening revealed innovative ideas that may hold relevance for the dairy industry, including whey valorization. For example, β-galactosidase immobilization on 3D-printed scaffolds for flow bioreactors (Shao et al., 2023), development of anti-adulteration detection tools using 3DP (Livas et al., 2021), production of innovative food packaging from biopolymers via 3DP (Wang et al., 2025), and creation of aerogels for tissue regeneration (H. Liu et al., 2024) represent notable developments.

CONCLUSION

The results of this scoping review confirm the significant potential of whey proteins in the development of 3DP materials. The key factor for success in this field remains the transition to practical applications. So, it is particularly relevant to further research studies on optimizing printing ink formulations.

To systematize accumulated knowledge, we propose the creation of a specialized database, based on the principles of physical colloid and food chemistry, which form an understanding of the beneficial interactions between different components. This tool will allow ingredients to be classified according to their functional properties, simplifying the development of new formulations. A promising trend is the development of predictive modeling tools to select ingredients, which can simplify formulation design based on a number of properties (rheological, textural, microstructural, and molecular interaction). The use of whey proteins beyond the food sector, particularly in biomedical scaffolds, may serve as a driver for the development of high-tech materials. Of particular interest is the integration of other whey protein ingredients, namely hydrolysates, into 3DP material formulations, which aligns with current global trends of research in the field of by-products. This review may serve as a methodological basis for research groups working with whey, offering strategies for planning experiments, analytical method selection, and innovation generation. The realization of these objectives could boost the development of competitive technologies across food and related industries.

AUTHOR CONTRIBUTIONS

Ekaterina I. Bolshakova: methodology; investigation; writing — original draft; visualization.

Natasha Poklar Ulrich: supervision; conceptualization; data curation; validation; writing — review & editing.

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