



Structuring Food Products Using 3D Printing: Strategies, Applications, and Potential

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Accepted: 17 October 2023

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Abstract

Purpose of Review It is the aim of this work to present different 3D printing structuring strategies used for customized food development, focusing on current high-interest trends such as encapsulation of bioactives, reduction of sugar, salt and fat, and production of meat analogues. Furthermore, we provide a critical discussion on future challenges and opportunities in this field.

Recent Findings While the most commonly used 3D printing technology in food production is layer-by-layer extrusion, it is also possible to create diverse structures by adjusting ink composition and process parameters, using co-axial nozzle assisted or dual extrusion, and employing different post-processing techniques.

Summary This review explores advanced strategies for food structuring via extrusion 3D printing that allow for customized food matrices, including texture, flavor, ingredient distribution, and encapsulation.

Keywords Food 3D printing · Food customization · Structured product · Meat analogue · Bioactive · Sugar-reduced

Introduction

Three dimensional printing (3DP), also referred to as additive manufacturing, involves the construction of an object layer-by-layer by a digitally controlled computerized process [1]. When applied to the food industry, the many benefits of this technology offer great potential for the development of new products. A key benefit of food 3DP is its capacity to produce intricate and personalized designs that are challenging to achieve through traditional manufacturing processes [2]. Furthermore, food can be tailored to meet individual preferences, such as shape, texture, flavor, and aesthetics. This level of customization elevates the overall eating experience, leading to increased consumer satisfaction. Moreover, food 3D printing plays a significant role in personalized nutrition, as it enables the creation of precise

food products with specific nutrient levels and functional compounds designed for addressing individual dietary needs and preventing health issues [1].

Various methods of 3DP have been explored for food production, including selective laser sintering, melting/soft-material extrusion, binder jetting, and inkjet printing [3]. Among these, the most widely adopted approach is extrusion-based 3DP (3DP-EXT). This technique fabricates 3D models by extruding fluid or semi-fluid mixtures (such as pastes or gels) and depositing them layer-by-layer on a platform. Notably, one of its greatest advantages lies in the extensive variety of raw materials that can be utilized to create food products [4].

The 3DP-EXT process begins with the creation of a computer-aided design (CAD) file in stereolithography (stl) format, to define the geometry and dimensions of the 3D object. This resulting stl file is then input into a slicing program, where various printing parameters are configured. These parameters, including layer thickness, infill, flow rate, layer height, print speed, and more are of utmost importance, as they directly influence the precision of the shape, internal structure, and overall quality of the final product [5]. The “ink”, which is formulated using food ingredients, is then loaded into a syringe, and subsequently extruded through a nozzle, driven by the force generated by a piston, screw,

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or air pressure. Subsequent layers are deposited by guiding the syringe to predetermined points specified by the CAD model. Depending on the type of material, various bonding mechanisms can come into play such as layer stacking controlled by the rheological properties of the materials, solidification through cooling, or extrusion of materials that form a gel.

Particularly, the ability of 3DP to deposit materials in a highly controlled geometric distribution and with certain patterns opens up several exciting possibilities: (1) encapsulation of bioactive components, alone or in combination, in different food matrices, allowing the preservation of their biological functionality and/or the control of their release; (2) precise distribution of ingredients in specific areas of food products, such as salt or sugar, to boost their flavor while using fewer amounts, resulting in a healthier food; (3) customization of food textures, very important in applications like the development of meat analogues with similar appearance and chewing properties to traditional meat. Therefore, this review focuses first on the strategies that have been explored to develop structuring products by using 3DP-EXT. Second, the most recent advances regarding the three mentioned applications are addressed to show the potential of this novel technology. Finally, future challenges and opportunities in this field are critically discussed.

Structure Engineering Through 3DP

Layer-by-layer construction in 3DP introduces unique textural characteristics compared to conventionally molded products. This distinction arises from 3D printing's capacity to generate structural differences at both macro and micro levels [4–7]. For example, for materials that undergo a phase change during printing, transitioning from a molten state in the nozzle to a semisolid state on the platform, layer-by-layer construction results in products with significantly different hardness compared to those produced using traditional molds [6]. Beyond the layer-by-layer effect, it is also possible to create diverse structures within a printed food matrix by adjusting various process parameters, ranging from simple to more complex, as we will explain below.

Printing Material Formulation

One of the easiest ways to create products with different matrix structures is by modifying the ink composition. In this case, the entire food product is printed with the same ink. By adjusting the ingredients, such as varying the types and ratios of fats, proteins, carbohydrates, or other components, it is possible to influence the texture, flavor, and even nutritional content of the final product. This level of customization enables the production of a wide range of matrix

structures, from soft and porous, to dense and homogeneous, all within the same printing process. As an example, Fig. 1a illustrates that clear differences can be observed in the appearance of cubes printed using inks containing two different concentrations of rice protein.

3D Printer Parameters

Modifications of 3D printer parameters offer a powerful means to engineer diverse matrix structures in printed objects. By selecting specific infill and pattern designs, it is possible to control the internal arrangement of material in a way that different degrees of solidity or porosity can be achieved. While the infill allows defining the level of solidity of an object, the pattern design provides additional versatility and allows the creation of matrices ranging from honeycomb-like lattices to triangular or rectilinear grids, enabling a wide spectrum of functional structural possibilities in 3DP. As an example, Fig. 1b shows printed samples of pumpkin puree with different structures obtained through changes in pattern design and infill.

Post-Processing

Certain 3D-printed foods may require a post-processing stage to achieve a stable and suitable condition for consumption. This critical step can involve various procedures, including cooking, cooling, or the removal of moisture through methods such as oven-drying or freeze-drying. Therefore, an additional strategy for achieving matrices with variable structures is the modification of post-processing conditions or methods, offering further flexibility in tailoring the final product's properties to meet specific requirements and preferences. Figure 1c shows the effect of freeze-drying—in this case applied to all samples under identical conditions—along with changes in ink formulation. By varying the amount of palm kernel stearin in high internal phase Pickering emulsions (HIPPEs) and applying freeze-drying to the printed products, microstructures with different pore sizes were obtained [8••].

Dual Extrusion

Dual extrusion enables the creation of diverse matrix structures through distinct layering and ordering of materials. By using two different extruders, each loaded with a different material, it becomes possible to alternate between them during the printing process. This method allows precise manipulation of material placement, enabling the creation of intricate matrices or even encapsulation when combined with complex design patterns. As examples, Fig. 1d shows a schematic representation of chocolate samples with different structures and total sugar composition obtained by

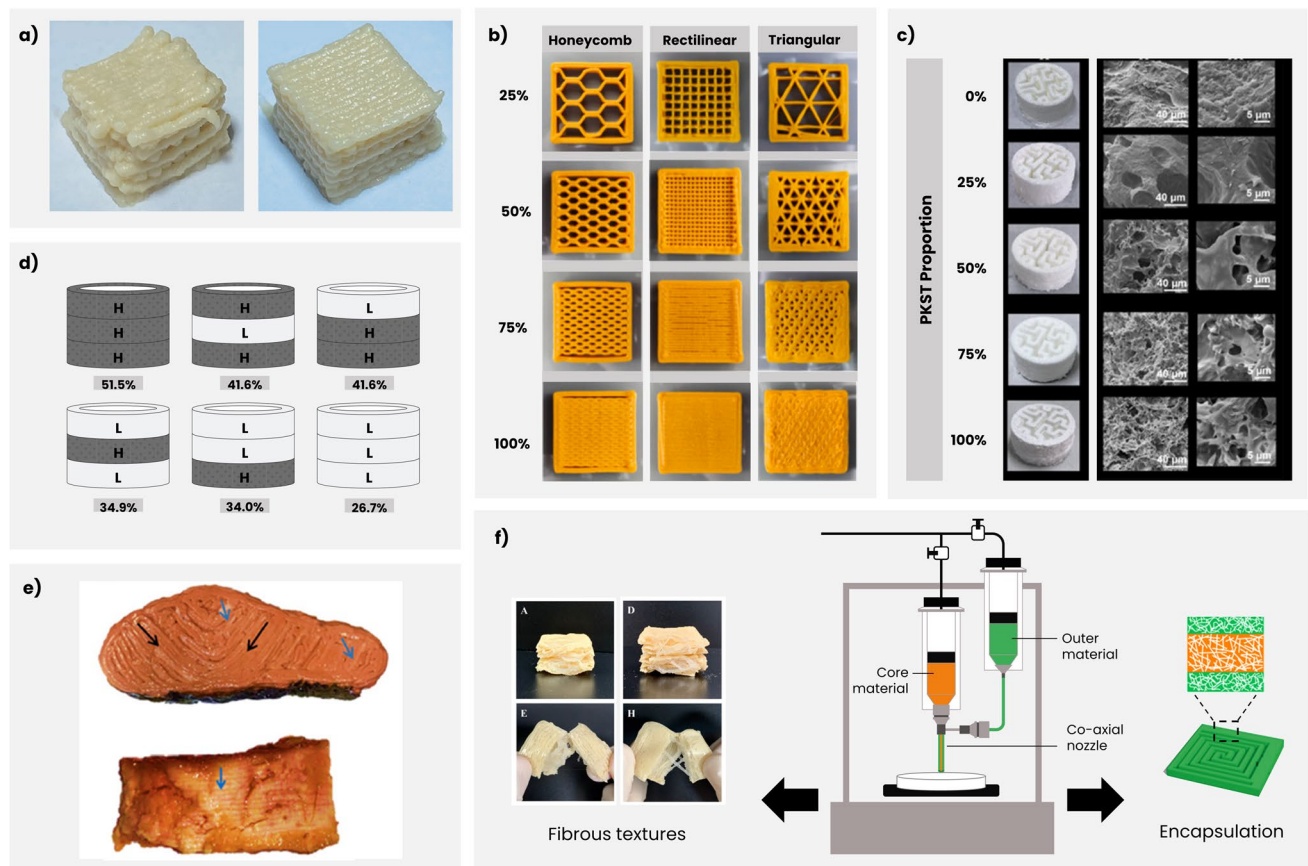


Fig. 1 **a** Effect of formulation on product appearance (reproduced from [2] with permission from Elsevier), **b** pumpkin puree printed samples obtained with different pattern designs and infills (reproduced from [50] with permission from Elsevier), **c** printed shapes using different proportions of palm kernel stearin (PKST) in oil and associated cryo-SEM images after freeze-drying post-processing (adapted from [8••] with permission from Elsevier), **d** cylinder sample designs for printing alternating high sugar layers (H, 51.5%) and low sugar layers (L, 26.7%) for reducing total sugar content

varying the layering order of printing materials (material I: high-sugar chocolate; material II: low-sugar chocolate), while Fig. 1e shows the use of dual extrusion to mimic the muscular characteristics of the meat in a salmon filet.

Co-axial Extrusion

Co-extrusion, facilitated by a specialized co-axial nozzle, is an ingenious 3DP technique that offers remarkable versatility in producing diverse and intricate matrix structures. In this method, a primary material is extruded through the inner nozzle, while a secondary material is simultaneously extruded through the outer nozzle. As it can be seen in Fig. 1f, this synchronized dual-material deposition can be used for applications in creating fibrous textures for meat analogues, as well as for encapsulation purposes. In the latter, sensitive ingredients requiring protection can be

(reproduced from [29••] under the terms of the Creative Commons CC-BY- 4.0 license), **e** top and side views of 3D printed salmon filet mimic achieved by alternating different ink compositions by dual-extrusion (black arrows indicate regions with myomere domain and blue arrows with myosepta simulant) (adapted from [45••] with permission from Elsevier), **f** schematic diagram of co-axial 3DP-EXT for ingredient encapsulation (adapted from [9•] with permission from Elsevier) or fibrous texture generation (adapted from [48•] with permission from Elsevier)

seamlessly incorporated into the core material, while a high-strength material serves as the outer layer. Thus, without altering the appearance of the 3D-printed product, the integration of components through a co-axial system enables the benefits of shape retention and encapsulation [9•].

Food Structures that Incorporate Bioactive Components

Bioactive components such as natural colorants, plant extracts (phytosterols, flavonoids, carotenoids, and plant polyphenolic compounds), prebiotics, and probiotics are well-known for their great health benefits, such as anti-cancer, antioxidants, anti-inflammatory, and anti-microbial properties [10]. In this sense, one of the most interesting and challenging applications of 3DP is the ability to incorporate

these bioactive minor ingredients into the ink's formulation preserving or even enhancing their functionality and bioavailability.

The literature on 3DP of materials incorporating bioactive components has significantly grown in the last 4 years (from a total of 5 documents in 2018 to 60 documents in 2023, search conducted in Scopus using the key words "bio-active*" AND "3D printing" AND "food" on September 4, 2023). In this review, we focused on the most relevant and recent publications that incorporate the following bioactive components: phytosterols, probiotics, polyphenols, carotenoids, and curcuminoids. Table 1 summarizes the main findings, stressing the strategies for the development of 3D printed structures and ink formulation.

Regarding structure engineering, Ahmadzadeh et al. [9•] used a co-axial nozzle for increasing stability of lutein by printing the core of the thread with a lutein and ethylcellulose solution, surrounded by a starch suspension on the outside. They found that this strategy significantly increased lutein retention index after storage compared to the physical mixture of all compounds. Similarly, Jeon et al. [11] produced starch-xanthan gum gels loaded with a nanoemulsion of curcumin by extrusion printing using a co-axial nozzle. The curcumin-loaded gels were the core ink, surrounded by an outer protective layer of bean paste. The authors proved, prior to the 3DP process, that the curcumin-loaded gel successfully retained its antioxidant activity. Interestingly, Jiang et al. [8••] combined ink formulation with printing post-processing strategies to adjust strength, porosity, and generate a shape response to stimulus in printed solids that incorporated phytosterol nanoparticles in a HIPPE. In order to obtain a material with a certain porosity, the object, constructed layer-by-layer by 3DP-EXT, was freeze-dried after printing to remove water. With the aim to generate a response to stimulus (also known as 4D printing), the printed object was exposed to a thermal stress that gave the desired geometrical deformation by gradual softening of the structured oil phase. Furthermore, the developed HIPPE loaded with phytosterol was tested for partial replacement of cocoa butter in preparing chocolate products, yielding promising results.

On the other hand, most of the contributions have focused on formulating printing materials with the aim to develop suitable structured matrices able to protect bioactive compounds from degradation and/or improve their bioavailability. In general, the most common formulation for 3DP inks are Pickering emulsions (PEs), since they offer the opportunity to tailor rheological properties to withstand the 3DP process. In addition to the work by Jiang et al. [8••], several other recent studies have explored the use of PE-based printing materials. Xu et al. [12] encapsulated *Bifidobacterium lactis* in the oil phase and used xanthan gum and tea protein as stabilizers, demonstrating that encapsulation improves the

viability of probiotics in the inks against heat treatment and digestion. Storage after 3DP as well as nozzle diameter did not show a decrease in viability, while keeping the material in the printer at high temperature (65 °C) for periods longer than 10 min highly decreased probiotic activity. Mohammadi et al. [13] formulated PE gels of canola oil and suspensions of different polyphenols, individually grafted onto soy protein isolate particles, to stabilize the emulsion and develop a 3D printed plant-based cheese. Tan and co-workers [14–16] formulated HIPPEs with astaxanthin in corn oil or algal oil and different stabilizers to provide stability and improve the bioaccessibility of this carotenoid, which was tested in the formulated inks. Liu et al. [17] and Huang et al. [18] incorporated β -carotene into sunflower oil or medium-chain triglyceride oil with different stabilizers verifying the protective ability of the encapsulation and the improved bioaccessibility of the formulated inks and their printability.

Traditional emulsions and nanoemulsion have also been used as printing materials for bioactive delivery. In this regard, Jeon et al. [11], Kavimughil et al. [19], and Leena et al. [20] managed the co-delivery of curcumin and resveratrol incorporated in gelatin hydrogels in an emulsion-based matrix, finding a synergistic effect of the developed inks for improving both bioactive bioaccessibility and intestinal target delivery evaluated in the 3D printed products [19] or in the formulated inks [20].

Oleogels and hydrogels, alone or combined as bigels, are also promising matrices for bioactive delivery. Cotabarren et al. [6] and De Salvo et al. [5] developed oleogels for phytosterol delivery that were successfully printed into stable and uniform solid forms. *Bifidobacterium lactis* and *Lactobacillus acidophilus* were encapsulated in alginate-gelatin hydrogels by Kuo et al. [21], then printed and freeze-dried to test the viability of probiotics during the entire 3DP process. Bigels enriched with quercetin (lipophilic polyphenol) and catechin (hydrophilic polyphenol) were developed by Xie et al. [22], proving that formulation greatly influences bigel structure and bioactive delivery of the formulated inks. Also, Zeng et al. [23] improved the printability of rice starch-based gels by incorporating catechin and procyanidin.

Direct incorporation of bioactive compounds in food-based matrices was tested by Liu et al. [24], who optimized a mashed potato formulation with xanthan gum and kappa-carrageenan to incorporate *Bifidobacterium animalis* and investigated probiotics viability in the printed products. Similarly, Yoha et al. [25] developed a flour encapsulating *Lactiplantibacillus plantarum* in a whey protein-maltodextrin matrix and evaluated probiotic viability before and after the 3D printing process as well as for different post-treatments; and, more recently, Cai et al. [26] formulated a 3D printed ready-to-eat food based on custard cream that co-encapsulates *Lactobacillus plantarum*, epigallocatechin gallate, and resveratrol proving a synergistic effect

Table 1 Recent studies on 3D printed structured products incorporating bioactive components, reduced sugar, salt, and fat content, and meat analogues

Printing material formulation	Objective	Strategies for structure development	Highlighted results	Reference
Bioactive compounds				
High internal phase pickering emulsions (HIPPE). Aqueous suspension of κ-carrageenan and <i>phytosterol</i> nanoparticles with soybean oil alone or incorporating palm kernel stearin	Develop a 3D printing matrix material with adjustable strength and pore size depending on the formulation and post-processing	Layer-by-layer 3DP-EXT varying ink composition. Pore generation: freeze-drying after printing. 4DP: geometrical deformation by thermal stress after printing	Formulation allowed unique thixotropy and structural recovery HIPPE low oil content enabled fat-reduced foods by 3DP and 4DP	[8••]
High oleic sunflower oil, monoglycerides, and <i>phytosterol</i>	Production of nutraceutical oral forms incorporating phytoosterols in an oleogel matrix	Layer-by-layer by 3DP-EXT varying ink composition and printing parameters (nozzle diameter, flow, extruder speed, and build platform temperature)	Stable and uniform solid forms with high weight repeatability Hardness affected by all evaluated parameters	[5, 6]
Different formulations of custard cream, <i>Lactobacillus plantarum</i> (free, encapsulated with gelatin (GE) and Sodium caseinate (Cas), or co-encapsulated with GE-Cas and EGCG), <i>Epigallocatechin gallate</i> (EGCG) (hydrophilic), and <i>resveratrol</i> (RSV) (lipophilic, in oil phase)	Develop a 3D printed ready-to-eat food co-encapsulating hydrophilic and lipophilic bioactives as well as probiotics. Evaluate formulation on: survival of probiotics after storage, heating, and gastrointestinal conditions; ink rheology; and printing properties	Layer-by-layer by 3DP-EXT varying ink formulation. Constant printing parameters	Co-encapsulation of EGCG and RSV with probiotic improved viability Custard cream exhibited gel-like rheological behavior and good printing properties, not affected by the addition of bioactives and probiotics	[26]
Pickering emulsion (PE) gels stabilized with xanthan gum and tea protein including <i>Bifidobacterium lactis</i> in olive oil	Feasibility of encapsulating probiotics in PE. Effects of 3DP parameters and storage on the viability of probiotics	Layer-by-layer by 3DP-EXT varying nozzle diameter and printing temperature	Encapsulation of probiotic increased viability against heat treatment, gastrointestinal digestion, and storage after 3DP. Nozzle diameter did not affect viability, T > 65 °C significantly reduced it	[12]
<i>Bifidobacterium lactis</i> and <i>Lactobacillus acidophilus</i> encapsulated in alginate-gelatin hydrogels	Develop hydrogel-based probiotics integrating encapsulation, 3DP-EXT and freeze-drying of the printed products to obtain shelf-stable food formulations	Layer-by-layer by 3DP-EXT with constant printing parameters and a product freeze-drying. Hydrogels formulation was varied	Freeze-drying allowed a solid-like state, increasing hardness and decreasing water activity Reasonable viability of probiotics during all the manufacturing process and after 4 weeks of storage	[21]
High-fiber high-protein composite flour matrix and <i>Lactiplantibacillus plantarum</i> encapsulated in a fructooligosaccharide-whey protein-maltodextrin matrix	Evaluate probiotics stability under different encapsulation techniques, 3DP process, and post-processing methods	Layer-by-layer by 3DP-EXT with constant printing parameters. Inks with encapsulated probiotics by spray, freeze, spray-freeze, and refractance window drying Different post-treatments (freeze, refractance window, hot air, and microwave drying)	No significant loss of probiotic viability during the 3DP process. Spray-freeze-dried encapsulation and freeze-drying post-processing showed the best survival rate and viability under in vitro digestion	[25]
<i>Bifidobacterium animalis</i> and mashed potatoes with xanthan gum and kappa-carrageenan	Optimize rheological properties for 3DP by formulation. Evaluate printing parameter effects on 3D printed products and probiotic survival. Evaluate storage conditions on probiotics viability and product appearance	Layer-by-layer by 3DP-EXT varying nozzle diameter, printing temperature, and holding time	Mashed potato formulation highly affected its extrusion behavior. Low nozzle diameter affected viability of probiotic. T > 55 °C reduced viability while storage did not affect bacteria survival	[24]

Table 1 (continued)

Printing material formulation	Objective	Strategies for structure development	Highlighted results	Reference
Oleogel: high oleic sunflower oil, mono-glycerides or lecithin, candelilla wax, and <i>quercetin</i> . Hydrogel: gelatin, water, and <i>catechin</i>	Develop printable bigels that deliver hydrophilic and lipophilic bioactives, explore different emulsifiers and different oleogel:hydrogel ratios. Evaluate release of bioactives by gastrointestinal digestion	Layer-by-layer by 3DP-EXT with constant printing parameters and different ink formulations	Oleogel:hydrogel ratios and emulsifiers had great influence on the bigels structure and the release of quercetin and catechin during simulated digestions	[22]
Pickering emulsion (PE) gels of canola oil and suspensions of <i>rosemary</i> , <i>thyme</i> , and <i>basil polyphenols</i> grafted onto soy protein isolate	Produce a printable PE gel incorporating different polyphenols to develop 3D printed plant-based cheese	Layer-by-layer by 3DP-EXT with constant printing parameters and different ink formulations	Ink formulated with soy protein-rosemary polyphenols displayed a 3D printed cheese analogue with improved lubrication, higher creamy sensation, and mouth-coating feature	[13]
Gel of rice starch, <i>catechin</i> , and <i>procyandin</i>	Improve printability of starch-based materials by incorporation polyphenols	Layer-by-layer by 3DP-EXT with constant printing parameters and different ink formulations	Polyphenols improved the extrudability of the starch gel	[23]
<i>Rose pollen (anthocyanin)</i> , yam paste	Evaluate effect of drying method (hot air, microwave vacuum, and freeze drying) on post-processing stability and quality of 3D printed rose-yam paste	Layer-by-layer by 3DP-EXT with different infill percentage	Microwave vacuum-dried printed products showed the best shape and color stability, high retention rate of bioactive substances, best flavor quality, and shortest drying time	[27]
High internal phase pickering emulsions (HIPPE) with <i>astaxanthin</i> in corn oil or algal oil and different stabilizers: gliadin/gelatinized starch nanocomposites, sea bass protein microgel particles, sea bass epigallocatechin-3-gallate	Formulate stable HIPPEs for astaxanthin delivery and 3D printing use	Layer-by-layer by 3DP-EXT with constant printing parameters and different ink formulations	Extrudability, printing performance, and self-supporting properties of the developed HIPPEs was confirmed	[14–16]
High internal phase pickering emulsions (HIPPE) with <i>β-carotene</i> in sunflower oil or medium-chain triglyceride oil and different stabilizers: walnut protein isolates (WPI), zein, tannic acid, and sodium alginate composites (ZTS)	Formulate stable HIPPEs for <i>β-carotene</i> delivery and 3D printing	Layer-by-layer by 3DP-EXT with constant printing parameters and different ink formulations	Stability and bioaccessibility of astaxanthin were significantly improved	[17, 18]
High amylose corn starch, ethyl cellulose, and <i>lutein</i>	Increase stability of lutein by encapsulation in starch-ethyl cellulose gels using 3DP with a co-axial nozzle setup	Layer-by-layer by co-axial 3DP-EXT (starch suspensions outside, lutein and ethylcellulose solutions inside) varying inks formulations, layer height, and printing temperature of starch extruder	WPI stabilized HIPPEs showed protective ability for <i>β-carotene</i> with good 3D printability ZTS stabilized HIPPEs' digestion showed higher release of free fatty acids and bioaccessibility of <i>β-carotene</i> with good precision and stability for 3DP	[9]
<i>Curcumin</i> and <i>resveratrol</i> stabilized in gelatin hydrogels with zein and polyethylene glycol (PEG) or ethyl cellulose (EC) nanoparticles, or emulsion-based matrix with gelatin and gellan gum and a medium-chain triglycerides oil	Improve bioavailability of synergistic bioactives by co-delivery in hydrogel or emulsion-based matrices	Layer-by-layer by 3DP-EXT with different ink formulations, printing speed, and nozzle diameter	Higher lutein retention in 3DP products after storage than non-encapsulated physical mixtures Zein-PEG and zein-EC nanoparticles improved bioaccessibility of curcumin and resveratrol Formulated emulsion protected curcumin and resveratrol of gastric conditions, allowing intestinal delivery. Matrixes able to be printed	[19, 20]

Table 1 (continued)

Printing material formulation	Objective	Strategies for structure development	Highlighted results	Reference
Nanoemulsion of <i>curcumin</i> , medium-chain triacylglycerol oil, Tween 20, and water in a gel of xanthan gum and potato starch	Produce starch-xanthan gum gels filled with nanoemulsions for customized food production, based on co-axial 3DP	Layer-by-layer by co-axial 3DP-EXT (bean paste outside, nanoemulsion filled gel inside) varying xanthan gum content	Nanoemulsion-filled gel physical properties, antioxidant activity, and printing performance varied with xanthan gum content	[11]
Reduced content of sugar, salt, and fat				
Dark chocolates with different <i>sugar</i> content: high sugar (51.5%) and low sugar (26.7%)	Investigate how layering chocolates with different sugar content influences the sensory profile, sweetness perception, and overall acceptance of 3D printed chocolates	Layer-by-layer by dual 3DP-EXT varying layering order of chocolate types	3DP to layer high and low sugar chocolate allows for a reduction in sugar content of up to 19% without changing the overall sweetness perception and overall liking	[29•••]
Chocolate pastes in which a portion (25, 50, and 75%) of the <i>cocoa butter</i> is replaced by arabic gum-based water-in-oil emulsions. Ratios of water to oil: 2:8, 3:7, and 4:6	Develop fat-reduced 3D printed chocolates with good printability	Layer-by-layer by 3DP-EXT with constant printing parameters and varying ink formulations	Successful 3D printed fat-reduced chocolates that maintained the polymorphic form of cocoa butter and excellent printing performance, with up to 75% cocoa butter substitution	[32]
Material composed of hydrated wheat starch and egg white powders with regular (6.5%) and reduced (3.3%) <i>salt</i> (NaCl) content	Structure matrix textures in starch-based material systems with spatial localization of sensory-active compounds, particularly NaCl	Layer-by-layer by dual 3DP-EXT of salted and unsalted layers	All printed structures, despite varying sodium localizations, exhibited comparable textural properties at the same infill levels. Inhomogeneous spatial distribution of NaCl-enhanced saltiness	[30]
Frozen silver carp surimi + 3% <i>NaCl</i> (Control) or its substitutes: 2.50–0.05% NaCl+0.32–1.58% CaCl ₂	Explore the impact of CaCl ₂ as a sodium replacement on the physicochemical attributes of silver carp surimi	Layer-by-layer by 3DP-EXT with constant printing parameters and varying ink formulations	Mixtures of NaCl with 0.32 and 0.63% CaCl ₂ resulted in increased hardness, improved rheological properties, and enhanced 3D printing capabilities of surimi gels	[36]
Water-in-oleogel emulsions (water fraction, (ϕ : 0.40–0.70). <i>Oleogel</i> with sunflower wax (SW, 1.0%) and variable soy-bean phosphatidylethanolamine (SP, 0.5–3.0%)	Investigate the use of water-in-oleogel emulsions for 3D printing fat-reduced foods, with a focus on improving printing performance and shape retention, and creating intricate designs	Layer-by-layer by 3DP-EXT with constant printing parameters and varying ink formulations	Optimal emulsions were achieved with ϕ = 0.65, 0.5 wt% SP, and 1.0 wt% SW. Increased SP content improved printing by enhancing viscosity, recovery, and self-support ability, resulting in precise hollow designs without collapsing	[33]
Oil-in-water emulsion with 90% aqueous sodium caseinate and <i>canola oil</i> (10%) or its reduced fat versions, where 15, 30, 45, and 60% of canola oil was replaced by acetylated microcrystalline cellulose (AMMC) stock suspension	Investigate the feasibility of using a reduced-fat casein-based Pickering emulsion stabilized by AMMC for 3D printing of cheese analogues	Layer-by-layer by 3DP-EXT with constant printing parameters and varying ink formulations	Incorporation of AMMC resulted in a more uniform and porous matrix. Varying AMMC solution levels had significant influence on morphology, printing performance, textural properties, structural characteristics, tribological behavior, thermal properties, and sensory attributes of the printed casein/oil matrix	[34]

Table 1 (continued)

Printing material formulation	Objective	Strategies for structure development	Highlighted results	Reference
Soy protein-based emulsion containing <i>canola oil</i> and biosurfactant solutions formulated with acetylated starch, octenyl succinic anhydride starch, ethyl (hydroxyethyl) cellulose, and dodecyl succinylated inulin	Study the effect of partially replacing oil with hydrophobically modified biosurfactants in a soy protein-based emulsion designed for the production of 3D printed meat analogues	Layer-by-layer by 3DP-EXT with constant printing parameters and varying ink formulations	3D printed meat analogues formulated with specific biosurfactants exhibited finer resolution, porous microstructures, increased hardness, and reduced friction coefficients, contributing to improved texture and sensory attributes	[35]
Meat analogues <i>Pea protein</i> , <i>soy protein</i> , <i>wheat protein</i> , coconut oil, <i>mushroom</i> xanthan gum, and potato starch	Develop and characterize a printable ink, analogue to meat, composed of vegan protein sources, enriched with mushrooms and with a higher amino acid content	Layer-by-layer by 3DP-EXT varying nozzle height, printing speed, and flow compensation	Nozzle height and printing speed were influential factors on precision and smoothness of prints Mushroom reduced hardness, stiffness, elasticity and chewiness; increased juiciness and nutritional value	[38]
<i>Soy protein (SP)</i> , <i>chickpea protein</i> , <i>potato protein</i> , <i>canola protein</i> , methylcellulose, and canola oil	Investigate 3D printing inks with new protein isolates for the development of meat analogues and compare with soy protein	Multilayer blocks construction by 3DP-EXT using inks of equal hardness with varying water-protein ratios	Chickpea protein could be used as a possible substitute for SP	[39]
Myomer simulant (orange muscle tissue): freeze-dried hydrated <i>red lentil protein</i> extract. Myosepta simulant (white adipose tissue): hydrated <i>yellow pea protein</i> extract. Both emulsified with camelina oil	Develop salmon imitations using vegetable protein-based food inks with a texture similar to salmon flesh	Layer-by-layer by dual 3DP-EXT with life-size dimensions. The myosepta simulant was printed by injecting it into the myomer simulant	A plant-based mimic with macronutrient content, physicochemical properties, and texture comparable to salmon fillet was developed	[45••]
<i>Soy protein (SP)</i> , xanthan gum (XG), and rice starch	3D printed fish analogues developed with edible soy protein-based inks	Layer-by-layer by 3DP-EXT, varying SP:XG ratio, nozzle diameter, and XY moving speed	Analogues of fish flesh with similar textural properties to the real yellow croaker meat. SP:XG ratio determined the printing accuracy	[43]
<i>Soy protein</i> , <i>wheat gluten</i> , <i>rice protein</i> , and canola oil	Investigate the rheological properties and printability of food inks produced from vegetable proteins for the development of 3D printed meat analogues	Layer-by-layer by 3DP-EXT using both unheated and air-heated printing of edible inks	Adding sufficient rice improved ink printing quality, enabling good printability and self-support Without rice protein, extrusion from the nozzle was difficult	[2]
<i>Cricket (Gryllus bimaculatus)</i> and <i>soy protein (SP)</i>	Characterize meat analogues produced using inks based on mixing ratios of SP and cricket gel	Layer-by-layer by 3DP-EXT varying cricket fraction (filtrate, supernatant and pellet) and its concentration, using a rectilinear infill pattern	Supernatant fraction was unsuitable at any concentration. Low concentrations of pellet fraction allowed high resolutions and well-defined dimensional specifications	[47]
<i>Soy protein</i> , sunflower oil, beet juice extract, and microcrystalline cellulose (MCC)	Produce a low-fat 3D printed meat analogue	Layer-by-layer by 3DP-EXT varying the concentration of sunflower oil and grafted MCC	Increasing MCC concentration improves ink mechanical strength, resulting in fine resolution, smooth surface, and no deformation of printed shapes	[44]

Table 1 (continued)

Printing material formulation	Objective	Strategies for structure development	Highlighted results	Reference
<i>Wheat gluten</i> , shiitake mushroom powder, Tween-80, water, cocoa butter, starch (wheat, corn, and potato), and <i>protein (soy and pea)</i>	Formulate gluten and plant protein-based gel materials for 3D printing and investigate their viability in the production of meat analogues	Layer-by-layer by 3DP-EXT varying printing temperatures, cocoa butter concentration, and using different starches and proteins	Below cocoa butter's melting point, poor ink flow affected formability. Excess cocoa butter reduced formability. Soy protein and wheat gluten inks were most suitable for 3DP	[37]
Soy protein paste: <i>soy protein</i> , potato starch, and xanthan gum. Fiber solution: kappa- and iota-carrageenan, sodium alginate, and glucomannan	Investigate a potential technology for producing alternative meat with fibrous structure inserted	Layer-by-layer by co-axial 3DP-EXT (fiber solution inside and soy protein paste outside) using a rectilinear infill pattern and different fiber solution concentrations	An optimal formulation was obtained that forms a strong and stable gel with elastic strength comparable to beef, suitable for texturing meat analogues	[48•]
Frozen surimi paste and potato starch solution	Study the possibility of producing products with the filament-like structure of crab meat using co-axial 3D food printing	Layer-by-layer by co-axial 3DP-EXT using inks with different concentrations of potato starch (frozen surimi inside and potato starch solution outside) and different infill patterns (aligner-rectilinear, rectilinear, and concentric)	Starch content affected dimensional stability; too little led to poor definition and deformation in prints, too much caused printing issues. Aligned-rectilinear pattern resembled commercial crab meat structure	[49]

3DP-EXT extrusion-based three dimensional printing, 3DP three dimensional printing, 4DP four dimensional printing

in improving probiotic viability in the formulated ink. For another application, Feng et al. [27] evaluated the effect of different post-processing drying methods on shape, color stability, retention of bioactives, and flavor quality for 3D printed yam paste with rose pollen (anthocyanin).

Food Structures with Reduced Content of Sugar, Salt, and Fat

Reducing the content of salt, sugar, and saturated fats in food products has become an imperative in the realm of nutrition and public health. Excessive consumption of these components has been unequivocally linked to a variety of health issues, including but not limited to obesity, diabetes, and cardiovascular diseases [28]. While these ingredients play a vital role in enhancing the palatability, texture, and shelf life of various food items, the need for moderation and healthier alternatives cannot be overstated. Novel technological approaches such as food 3DP provide a promising pathway for the development of healthier food products. For instance, food 3DP offers the possibility of using reformulated mixtures as printing materials, in which harmful ingredients could be reduced, eliminated, or substituted with healthier counterparts, while maintaining similar physical and sensory properties to the original ones through the meticulous design of microstructures.

One of the most captivating approaches found in the literature for the development of foods with reduced sugar and salt content involves using 3DP to precisely control the spatial distribution of these ingredients within the entire food matrix (Table 1). With this aim, Khemacheevakul et al. [29••] conducted a noteworthy study exploring the production of hollow cylinder chocolate samples with different global sugar concentrations using a layer-by-layer construction technique that varied the layering order of high-sugar (51.5%) and low-sugar (26.7%) chocolate types. The authors conducted a sensory analysis to evaluate the effect of the layering design on consumer perception. The results showed that the use of 3DP for layering chocolates allows for a 19% reduction in sugar content without compromising the perceived overall sweetness and overall liking of the products. Furthermore, the study indicated that the order in which high and low-sugar chocolates were layered significantly influenced the perceived overall sweetness and temporal sensory profiles of the 3D printed chocolates. Similarly, Famhy et al. [30] explored the creation of structured matrices using different layering designs of starch-based food materials with both normal and reduced salt content. Through the innovative application of dual extrusion and on-board near-infrared heating, they developed a 3DP method that not only allows precise control over texturing but also enables the spatial distribution of sodium chloride. The study revealed that all

printed structures, regardless of varying salt localizations, maintained consistent textural properties at the same infill levels, and the inhomogeneous spatial distribution of sodium chloride led to an intensified perception of saltiness. This finding holds promise for the development of reduced-salt products with enhanced flavor perception. In a recent study [31], a sensory design approach involving layering was employed to create more appealing lemon mousses for hospitalized patients. The authors discovered that bilayer lemon mousses, featuring lower acidity on top and higher acidity on the bottom, consistently received higher liking and desire scores compared to mousses with uniform acid levels distributed in a monolayer. Moreover, the bilayer configuration resulted in a significant 13% increase in food intake among patients. While this study did not employ 3DP for layering, it suggests the possibility of strategically adjusting the acidity of sweet products to develop low-sugar versions that can achieve equal or even higher acceptability.

Another strategy that has been employed to create healthier reformulated products involves the use of inks formulated with variable and reduced content of specific ingredients, combined with 3DP technology, to build entire products layer-by-layer (Table 1). This approach has been mainly applied to develop 3D-printed foods with reduced fat content. In particular, the use of emulsions and oleogel-based emulsions as 3DP materials has been investigated to effectively reduce the fat content in food products. For example, You et al. [32] developed chocolate pastes by replacing different proportions (25, 50, and 75%) of cocoa butter with water-in-oil emulsions based on arabic gum. The findings revealed that it was possible to successfully replace up to 75% of cocoa butter with emulsions at specific water-to-oil ratios, resulting in chocolates that maintained the desired polymorphic form of cocoa butter and exhibited exceptional printing performance, yielding well-defined shapes for the printed objects. Similarly, water-in-oleogel emulsions were tested as printing materials, finding that those formulated with a high- water fraction (0.65) had the highest support ability and allowed producing more precise designs without object collapsing [33]. Shahbazi et al. conducted studies focused on reducing fat content in cheese and meat analogs while assessing their impact on printed product quality. Their research revealed that substituting canola oil with acetylated microcrystalline cellulose in cheese analogs resulted in transformed structures, enhancing printing performance and sensory attributes [34]. Additionally, by replacing canola oil with hydrophobically modified biosurfactants in soy protein-based emulsions designed to print meat analogs, they achieved improved texture and sensory attributes characterized by finer resolution, porous microstructures, increased hardness, and reduced friction coefficients [35]. Furthermore, a recent study adopted this approach to reduce salt content in silver carp surimi [36]. The researchers found

that substituting a portion of sodium chloride with calcium chloride not only reduced sodium but also led to increased hardness, improved rheological properties, and enhanced 3DP capabilities of surimi gels.

Meat Analogues Food Structures

Meat is an important part of the human diet, and its consumption is steadily increasing due to the world's growing population. In fact, according to the Food and Agriculture Organization of the United Nations (FAO), the global population is estimated to reach 9 billion people by 2050, resulting in 73% increase in demand for meat, including pork, poultry, and beef [37]. Likewise, plant-based diets have gained popularity in recent years, driven by environmental awareness, the quest for sustainability, the desire for a healthier lifestyle, and compassion for animals. This trend has led to rapid growth in the plant-based food industry to meet consumer expectations [38, 39]. An emerging focus in this area is on the creation of 3D-printed structured products known as meat analogues, which aim to replicate the flavor, texture, appearance, and nutritional value of traditional meat [40]. The scientific literature on 3DP of meat analogues is limited but growing, with approximately 30 papers identified in a Scopus search using the keywords “3D print*” AND “meat* analogue*” on September 7, 2023. In this review, we focus on the most relevant publications on meat analogues incorporating plant proteins, fungi and/or edible insects, which are summarized in Table 1.

Regarding ink formulation for meat analogues, these mixtures have generally been prepared with 50 to 80% of water, which is essential for the juiciness of the product [38, 41]. Proteins, varying from 15 to 25%, play multiple functions in the product's structure, including hydration, gelling, texturization, and more. Fat, up to 15%, contributes to softening, juiciness, flavor release, and mouthfeel. Flavorings (3% to 10%), binding agents such as xanthan gum and vegetable starch (1 to 5%), and coloring agents such as beet root extract (0 to 0.5%) have also been added [38, 41, 42]. The combination of these ingredients allows meat analogues with acceptable sensory qualities [38].

Soy protein has been widely adopted as the primary alternative protein source in the food sector, owing to its low cost, excellent nutritional content, and adaptability [39]. This trend has been also observed in food 3DP. Among authors who evaluated the printability of soy protein isolate (SPI)-based inks, we can highlight Shi et al. [43] who successfully printed fish analogues with a texture similar to real meats by varying SPI concentrations and optimizing printing parameters, and Shahbazi et al. [44] who replaced the oil phase of meat alternatives with soy-based PEs, preserving both the textural and sensory characteristics of the

products. Regarding the use of other plant protein sources, Qiu et al. [2] fabricated meat analogues by replacing part of the soy protein with rice protein and using a texturization technique based on air heating-assisted 3DP. Other researchers have shown that chickpea, lentil, pea, and rice proteins have potential to be used as an alternative to soy protein in 3DP-EXT [37, 39, 45••].

On the other hand, products containing edible mushrooms are more attractive and tastier than other plant-based foods, while present at the same time health benefits such as anti-inflammatory, immunomodulatory, anti-tumor, anti-diabetic, and functional relief from constipation [46]. In this sense, Demircan et al. [38] improved the textural properties and juiciness of 3D printed meat analogues by incorporating three different varieties of mushrooms, which increased their acceptability, nutritional value, and umami taste. More recently, alternative ingredients such as edible insects have been evaluated as promising meat sources. In fact, when compared to other alternatives, edible insects emit significantly less greenhouse gases and ammonia, have high feed conversion efficiency, short life cycles, and a low environmental footprint. With this aim, Nam et al. [47] investigated printable inks based on soy protein and different cricket fractions, concluding that the cricket filtrate fraction resulted in products with closest dimensions to the 3D design.

While most printed meat analogues have tailored the printing design to meet the texture of the real meat, new approaches have been developed to better replicate the filamentous characteristic. Ko et al. [48•] used co-axial nozzle-assisted 3D printing to produce meat analogues with a “real” texture by coating a fiber solution with a soy protein paste and determined the optimal formulation that allowed maintaining its shape, even after post-processing. Kim et al. [49] used a potato starch solution on the outside and surimi as core material to simulate crab flesh, exploring different infill patterns and concluding that the aligned-rectilinear design best resembled commercial crab meat. On the other side, Tay et al. [45••] used a dual extrusion technique to produce a salmon analogue. They first printed a material that represented salmon muscle and then injected, by 3DP as well, the adipose tissue simulant.

Conclusions

In this review, we have delved into the strategies for structuring food products using extrusion 3DP, highlighting the current and most advanced key methods for creating diverse matrix structures. These methods include modifying ink composition, adjusting 3D printer parameters, and using post-processing techniques, as well as dual and co-axial extrusion. Each of these strategies offers unique advantages for creating custom food structures, from adjusting texture

and flavor to precise ingredient distribution and encapsulation. In this section, we draw the primary conclusions, as well as future challenges and opportunities that arise from applying these strategies in three areas of high interest: the encapsulation of bioactives, the reduction of sugar, salt, and fat, and the production of meat analogues.

Regarding the incorporation of bioactive components into food matrices, we found that it can be addressed through two strategies. On one side, there is a need to develop edible materials that can withstand the 3DP without severe adjusting of the process temperature to preserve the beneficial activity and viability of the compounds. In this sense, some mashed vegetables and gelatinous substances can be used, but the strength of the printed objects is low which limits the application and has turned the attention to Pickering emulsions, hydrogels, oleogels, and similar structured materials. On the other side, tuning of the printing process itself can greatly enhance the performance of the bioactive components such as the use of co-axial nozzles that can encapsulate the sensitive materials to protect them from environmental degradation. The ability of the 3DP process to geometrically distribute inks as desired also opens up great opportunities for the design of patterns that can also meet this protective functionality or that can tune the release profile as desired, for example by using dual extrusion or multiple printing. These strategies need to be further explored for generating 3D printed products that can meet new ways of delivering bioactive substances. Another critical consideration is related to determinations of bioactive compound activity. To evaluate whether 3DP offers advantages over traditional manufacturing methods, it is essential to test bioactivity not only in the printing materials but also in the final printed products. Ideally, these assessments should also include comparisons with products manufactured using conventional techniques.

We have also identified a huge potential for food 3DP in the development of reduced sugar, salt, and fat products. By using 3DP for strategic layering and precise distribution of ingredients throughout the entire food matrix, it becomes possible to achieve healthier alternatives with sensory properties similar to the original products. Additionally, we have highlighted the potential of replacing fat with emulsions and oleogels to create low-fat 3D-printed foods while maintaining desirable sensory attributes. As a possibility, layering using dual extrusion together with changes in design patterns could be combined to develop even more complex matrices with optimal ingredient distributions and improved palatability.

Regarding the development of meat analogues, we consider that 3DP arises as the most promising technology for this type of product since it easily allows the construction of fibrous structures similar to real meat. However, this application is at a very early stage of development and requires a lot

of research to be cost-effective, as well as facing consumer acceptance challenges.

The opportunities presented by food 3DP in the quest for healthier, tastier, and more customized food products are undeniable. With ongoing innovation and collaboration across various disciplines, 3DP has the potential to revolutionize the way we approach nutrition and food security, offering consumers a broader range of healthier and equally enjoyable food choices.

Acknowledgements The authors express their gratitude to Dis. Teresa Dutari for her assistance.

Author contributions Ivana M. Cotabarren: conceptualization, investigation, writing—original draft preparation, writing—review and editing, supervision, funding acquisition; M. Itati De Salvo: investigation, writing—original draft preparation, writing—review and editing; Camila A. Palla: conceptualization, investigation, writing—original draft preparation, writing—review and editing, supervision, funding acquisition. All authors contributed to and approved the final draft of the manuscript.

Funding This work is financially supported by the Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT) through PICT-2021-GRF-TI-00273 and Universidad Nacional del Sur (UNS) of Argentina through PGI 24 M/163 and PGI 24/M172.

Declarations

Competing interests The authors declare no competing interests.

Conflict of Interest The authors declare no competing interests.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

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- Of importance
- Of major importance

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