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3D Printing of meat

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Abstract

Three-dimensional printing (3DP) process stands as a developing technology for food manufacturing, which offers the opportunity to design novel food products with improved nutritional value and sensorial profile. This review analyses the potential applications of 3DP technology for meat processing and the elemental aspects affecting the printability and post-processing feasibility of 3D printed meat products. The combination of nutritionally balanced ingredients and novel internal structures may be schemed into a multi-material 3D model that meets special individual needs, such as chewing and swallowing difficulties. Furthermore, a temperature-controlled extruder-type 3D printer built with multi-head system is suggested to suit the required conditions for meat safety and rheological requirements.

Keywords

3D printing; Meat products; Recombined meat; Food design

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1. Introduction

Only 7.2% in weight of a cattle carcass accounts for cuts that are considered suitable for high-value steaks (Conroy, Drennan, Kenny, & McGee, 2010). In order to obtain specific meat cuts based on customer specifications, trimmings and off-cuts with varying composition and quality are remained and often, either sold as low-value by-products, or even considered waste. According to Bonny, Gardner, Pethick, and Hocquette (2017), although it is unlikely to eradicate the conventional practices for meat production, in a near future unconventional protein sources are likely to represent an increasing competitive alternative for inferior meat cuts and processed meats made from meat by-products. This signifies an important amount of nutritious meat tissues that might be misused. Consequently, producers are continuously in the search of new technologies, such as restructuring meats or value-added cuts (Yeh,

Omaye, Ribeiro, Calkins, & de Mello, 2018) to increase profitability and global competitiveness.

An emerging technology for the food industry, which represents a great opportunity to seize meat by-products for the fabrication of customized meat products, is three-dimensional printing (3DP). 3DP technology uses a computer-aided design (CAD) software assisting a digital manufacture machine in the generation of three-dimensional objects without any additional tool (Noorani, 2017). Besides already standing as a relevant technology in the medical, automotive, aerospace and fashion fields (Gross, Erkal, Lockwood, Chen, & Spence, 2014), during the last decade, 3DP technology has also gained the attention of researchers in the food science field due to the potential advantages that 3DP could bring to the food industry in the future.

Based on the additive manufacturing (AM) process, which consists of a layer-by-layer deposition with predetermined thickness to create complex freeform structures (Noorani, 2017), 3DP offers the possibility of manufacturing novel food products with digitalized intricate shapes, inexperienced textures and higher nutritional value, through the combination of different food ingredients and printing methodologies. 3D printing methodologies applied to food ingredients are extrusion, inkjet printing, binding deposition, and bio-printing (Godoi, Prakash, & Bhandari, 2016). However, the former one is the most commonly used due to its suitability to a wide variety of food rheological properties.

The application of 3DP in the food science field comprises various aims such as novelty/fun/creativity, convenience and efficiency, health/nutrition, reducing waste and enhancing environmental sustainability, and alleviating world hunger (Turner & Lupton, 2017). For instance, one of its most relevant applications relies on the design of personalized food meals aimed for elderly consumers dealing with swallowing and/or mastication difficulties, developed as part of the PERFORMANCE project (RTDS Group, 2014).

However, in order to manufacture a 3D printed meat product with a desired design, sensorial profile and nutritional value, first the printability of the meat paste needs to be assessed. The printability of any food material refers to its ability to be handled and dispensed by a 3D printer into a freeform structure after deposition (Godoi et al., 2016), and is affected by the printing conditions and the rheological properties of the materials (Kim, Bae, & Park, 2017).

Based on the printability of food ingredients, three categories were identified by Sun, Zhou, Huang, Fuh, and Hong (2015): native printable food materials, non-native printable traditional food materials, and alternative ingredients (Figure 1).

- A material with native printability has enough flow ability to be easily extruded from the nozzle without additional flow enhancers (Sun et al., 2015). Some natively printable materials, including cream cheese, cheddar cheese (Kim et al., 2017), Vegemite and Marmite (Hamilton, Alici, & in het Panhuis, 2018) have enough rigidity to uphold its structure after deposition, and are thus suitable for sophisticated 3D objects and general 3D printing system. However, other materials may be easy to extrude but present difficulty to withstand a 3D structure, as is the case with Greek yogurt and ketchup, recommended for 2D printing only (Kim et al., 2017).
- On the other hand, non-native printable traditional food materials require additional flow enhancers for ease of extrusion and/or post-cooking processes (Sun et al., 2015). Most traditional staple foods lack printability characteristics, requiring aided and controlled rheological and mechanical behaviour during printing and deposition. Therefore, the effect of flow and viscosity enhancers on the printability of food materials have been widely studied. For example, Wang, Zhang, Bhandari, and Yang (2018) proposed a surimi gel by combining silver carp surimi with 1.5% NaCl as a food material suitable for printing 3D complex patterns. Also, Severini, Derossi, Ricci, Caporizzi, and Fiore (2018) added 1% of fish collagen to enhance the viscosity of a fruit and vegetable blend to successfully build edible pyramids. However, when adding agar to celery, Lipton, Arnold, Nigl, Lopez, Cohen, Norén, and Lipson (2010) obtained an extrudable celery fluid gel which was not able to hold 3D printed structures, although concentrations are not reported. Other additives commonly used for 3DP applications include gelatin, xanthan gum, starch, pectin and alginate (Vancauwenberghe, Katalagariakakis, Wang, Meerts, Hertog, Verboven, Moldenaers, Hendrickx, Lammertyn, & Nicolai, 2017).
- Alternative ingredients refer to those emerging as novel sources of functional constituents aiming to customize nutrition, such as proteins and fibres isolated from insects, algae, fungi, bacteria, among others (Sun et al., 2015). These alternative ingredients are becoming of interest as potential supplements towards a balanced nutrition, complementing traditional food sources, such as cattle and crops, and can be formulated into a paste or powder suitable for 3D printing within a meat paste for the production of customized meals. For instance, the combination of entomogaphy (eating insects) with

3DP technology has been tested by adding edible insect powder from *Tenebrio molitor* to enrich the protein content of 3D printed wheat-based snacks (Severini & Derossi, 2016b). Similarly, insect protein based flour made from mealworms, crickets and silkworm pupae was combined with food carriers such as icing butter, chocolate, cream cheese and spices to form an extrudable paste which was printed into insect's inspired shapes by Soares and Forkes (2014) as part of the Insect Au Gratin project. Yet, consumers' awareness and acceptance of these type of foods might present double the challenge: firstly, introducing the novel technology of 3D printing into their kitchens and secondly, the addition of insects into their diets.

Meat and its by-products are fibrous materials non-printable by nature (Liu, Ho, & Wang, 2018a), which require the modification of its rheological and mechanical properties through the addition of flow enhancers to obtain an extrudable paste-like material. To date, only few studies about 3D printing of fibrous materials, such as meat and seafood have been published. Lipton et al. (2010) assessed the suitability of 3D printed turkey meat added with transglutaminase (TGase) and bacon fat for conventional post-processing (sous-vide cooking). Likewise, Liu et al. (2018a) were able to 3D print chicken, pork and fish in a slurry form with the addition of gelatine solution. Also, the printability of fish surimi with added NaCl was assessed by Wang et al. (2018), whilst blended canned tuna with spring water was 3D printed as part of a meal designed for people with swallowing difficulties (Kouzani, Adams, Whyte, Oliver, Hemsley, Palmer, & Balandin, 2017). Additionally, during the 3D Food Printing Conference Asia-Pacific, Meat and Livestock Australia (2017) proposed the creation of meat scrolls made from emulsified secondary cuts, which well maintained their shape after frying.

Nonetheless, no published data exists regarding the printability of beef meat and thus comprehensive informative data concerning the desirable rheological and mechanical properties of the meat paste in order to be printed is still required, whilst considering the safety issues of meat products and the most suitable printing conditions.

2. 3D Printing process

Three-dimensional printing, also known as additive manufacturing (AM), is a process that generates freeform structures by introducing a prototype into a computer aided design (CAD) software, which is then converted into a .STL file by a slicing software to be recognised and processed by 3D printers (Noorani, 2017). The technology involves a layer-by-layer

deposition with predetermined thickness to create complex three-dimensional objects from different materials used as “inks”, using strictly the necessary amount of material to consolidate the shape of the printed object. It has become a relevant technology with broad applications in the medical field for tissue engineering, automotive and aerospace fields for components design, as well as fashion and food design, among others (Gross et al., 2014).

3DP offers an alternative technology with sustainability benefits such as reduced demand of raw materials, workforce, energy and transportation (Peng, 2016; Sher & Tutó, 2015).

However, some challenges remain to be resolved by the 3DP industry such as time consumption and initial inversion, limited printable materials, accuracy and surface finish (Noorani, 2017).

Most commonly used materials for conventional 3DP are polymers, metal, ceramics and their composites (Noorani, 2017), although food materials have also gained the attention of researchers during the last decade.

Through the combination of diverse food materials and 3D printing methodologies, the design of novel food products offering unique textures, nutritional value and eating experiences is conceivably unlimited. In this way, besides waste conversion through the added-value chain, the development of health and well-being products, as well as novel food interactions may be triggered. For instance, meat trimmings and off-cuts can be further processed into a paste and 3D printed with varying structural arrangements in order to make it suitable for dysphagia patients with chewing, swallowing and digesting difficulties. While, at the same time, allowing the design of personalized food and eating experiences through the targeted addition of supplementary ingredients.

However, in order to manufacture a printed food product with desired design and nutritional value, several aspects needed to be taken into account to ensure the required printing precision and accuracy. Some of these aspects, as reported in the literature, include but are not limited to the printing machines, methodologies, prototype design and software, food ingredients and additives, processing parameters, and post-processing suitability (Liu, Zhang, Bhandari, & Wang, 2017) applied to each 3D printed food manufacturing process.

2.1 Current application of 3DP in food products

In the last decade, 3DP technology for food products has increasingly developed through its application to a wide range of food materials. Chocolate (Mantihal et al., 2017), dough Severini et al. (2016a), cheese (Kim et al., 2017), fruits and vegetables blends (Severini et al.,

2018), hydrogels (Yang, Zhang, Bhandari, & Liu, 2018), and combined powdered food materials into balanced meals (Diaz, Van Bommel, Noort, Henket, & Briër, 2016) are some of the mostly studied food materials.

For 3D printed foods that do not require post-processing operations (e.g. cooking), such as chocolate and cheese, the studies commonly assess how the rheological and mechanical properties of food materials affect the printability (Lille, Nurmela, Nordlund, Metsä-Kortelainen, & Sozer, 2018; Vancauwenberghe, Verboven, Lammertyn, & Nicolai, 2018; Yang et al., 2018), and how the printing parameters affect the printing accuracy (Hao, Mellor, Seaman, Henderson, Sewell, & Sloan, 2010; Severini et al., 2016a; Yang et al., 2018). Nonetheless, few studies (Lipton et al., 2010; Lipton, Cutler, Nigl, Cohen, & Lipson, 2015; Severini et al., 2016a) have taken into account the post-processing feasibility of the 3D construct for materials such as dough or meat, which require further heat treatment; for instance, its ability to withstand cooking operations without losing the 3D intricate design due to cooking loss/shrinkage. In general, there is still an extensive field for research regarding the application of this technology for a broad range of foodstuffs with varying formulations.

2.2 3D Printing of meat

To date, only a small number of studies account for the printability of fibrous-meat materials, through the assessment of the rheological properties of the meat “ink”, as well as the post-deposition and post-processing properties of the printed object. Lipton et al. (2010) authored the first published work on 3D printing of meat. The authors demonstrated the suitability of 3D printed turkey puree for sous-vide cooking method. The turkey paste was added with TGase as a binder, and bacon fat as a flavour enhancer, and was printed in the shape of a truncated hemisphere (Figure 2) using a Fab@Home extruder- type 3D printer. Also, the same slurry was used to print a cube containing celery fluid gel inside. Likewise, gelatine was added to fibrous meats (pork, chicken, fish) as a viscosity enhancer (Figure 3) to evaluate its printability and the applicability of a newly designed 3D printer for fibrous materials (Liu et al., 2018a), although the post-processing viability was not assessed. Such introductory results in 3D meat printing show how this technology can further generate meat products with complex internal structure, containing on-demand functional ingredients and modified textures for enhanced eating experiences.

In addition, during the 3D Food Printing Conference Asia-Pacific, Meat and Livestock Australia (2017) proposed the creation of an emulsified red meat ink from secondary cuts,

which was 3D printed into meat scrolls with a ByFlow 3D printer that well-maintained their shape after frying. In addition, the printability of seafood materials has also been tested to some extent. Recently the printability of fish surimi gel was assessed by Wang et al. (2018) using a screw-conveyor extruder type 3D printer. The effect of added NaCl (0%, 0.5%, 1%, 1.5%) to fresh silver carp fillet mince on the functional and rheological properties of the surimi gel was evaluated through the water holding capacity (WHC), gel strength, rheological behaviour, microstructure and distribution of water content within the gel; suggesting 1.5% NaCl as the optimal concentration for having suitable mechanical properties for 3D printing process (Figure 4). Furthermore, the authors evaluated the effect of printer settings on the geometrical precision and dimension of the deposited structures, although no objective comparison was performed among printed structures, such as the post-deposition and post-processing properties.

Furthermore, aiming to develop pureed foods for people with swallowing difficulties, tuna puree was obtained by blending tuna in spring water for 5 minutes, and was further 3D printed into a tuna shape with pressure-controlled extrusion at 20 °C (Kouzani et al., 2017). Such processing temperature may compromise not only the material behaviour, but also the food safety risks, showing a limitation of 3D printers that still need to be addressed in order to be applicable for a wider variety of foods.

2.3 3D Food printers and printing parameters

The basic components of a 3D food printer stage include a motor-driven print-head and a platform, commonly attached to a stage with Cartesian configuration (Sun, Zhou, Yan, Huang, & Lin, 2018). Based on the 3D printing methodology built into the 3D printer, the print-head and platform characteristics may vary. For most 3D food printers, the print-heads are extruder-type, and single and dual nozzle models are presented in Figure 5. Extruder-type printers are suggested as the most suitable for meat paste printing, as described in Section 2.6. Some examples include the 3D Dual Nozzle model Shinnove-D1 (Hangzhou Shiyin Technology Co. Ltd.) suitable for multi-food materials, Porimy 3D Food Printer (Kunshan Bolimai 3D Printing Technology Co.) suitable for soft-food materials, as well as non-food machines, which have been adapted to suit food materials processing, as is the case of the Fab@Home printer which was used to print turkey meat and scallop (Lipton et al., 2010).

New 3D printing approaches are still under development aiming to overcome the existing challenges of suiting meat materials into the additive manufacturing process, so called Food

Layered Manufacture (FLM), which requires standardized flow and setting properties of food materials (Wegrzyn, Golding, & Archer, 2012). Some of the approaches still under improvement include a 3D meat printer with improved ink storage capacity and extrusion quality (Liu et al., 2018a) and the bio-printing process of engineered meat fibres from cultured stem cells (Forgacs, Marga, & Jakab, 2014).

Nonetheless, adjustments of 3D food printers are still required in order to fulfil meat-processing conditions, like storage capacity of the hoppers or cartridges, and temperature control during the printing process, on behalf of safety issues. Current 3D printers focus on the nozzle temperature, which is particularly designed to control the physico-chemical behaviour of limited types of materials, such as chocolate. However, when printing meat, the temperature should be controlled in an uninterrupted manner, throughout the feeding system, the hopper, the nozzle and the platform itself. With the intention of meeting adequate temperatures for perishable foods, both, the feeding system and the stage are suggested to maintain temperature-controlling devices in the range of freezing and cooling temperatures. For example, when printing meat and seafood products, it is crucial to maintain processing temperatures below 4°C to prevent microbial growth. However, some previous studies on meat and seafood printing focused on the extrusion and printing process (Kouzani et al., 2017; Liu et al., 2018a), post-deposition and post-processing conditions (Lipton et al., 2010), rheological and mechanical properties of the material (Wang et al., 2018), regardless of safety concerns during printing due to the printer's limitations. When a printer is not attached with cooling system, the suitability of the technology for the processing of highly perishable materials like meat is dependent on the initial meat paste temperature and the period of time that the meat paste remains in the cartridge or platform at ambient temperature. The later one is, in turn, dependent on various aspects, such as the volume of the cartridge vs. the volume of the printed design, the printing time, and the number of printed objects per cartridge, etc.

2.4 Printing conditions to enable 3D printing of meat

Several studies demonstrate the effect of varying printing processing parameters on the printability of food materials and hence, the quality of the final printed objects. The component settings that include nozzle speed, nozzle height (layer height), nozzle diameter, extrusion rate, and infill percentage, are suggested as critical parameters affecting the geometry accuracy of the printed construct (Hao et al., 2010). While different mechanical properties of the 3D printed meat paste can be obtained by varying the critical printing

parameters, each of them affect the accuracy of the printed geometry in individual and/or combined ways.

In this way, the selection of the nozzle diameter should take into account the desired accuracy of the printed structure and the food components within the meat paste. A nozzle diameter $>2\text{mm}$ may facilitate the extrusion of the paste containing bigger particle size components, such as connective tissue but the printing precision may be compromised by the deposition of thicker streams. Whereas a nozzle diameter $<2\text{ mm}$ allows the production of more accurate and intricate objects but the formulation may be compromised, as fine emulsion-like pastes are required for the extrusion through the narrow nozzle without the occurrence of clogging.

Similarly, an optimal nozzle height determines the accuracy and dimensions of the printed meat product, and it is suggested to be equivalent to the dimension of the nozzle diameter. Due to the extrudate swell phenomenon (Kim et al., 2017) attributed to the springiness of meat paste, a nozzle height lower than optimal may result in scattering of the deposited stream, thus producing expanded objects as compared to the desired design. Whereas the opposite situation, a larger than optimal nozzle height, may result in the dragging of the meat paste stream since it is not properly deposited on top of the former layers (Figure 6A), contributing to the void fraction within the structure which in turn, can affect the post-processing changes in the meat product. In addition, springiness of the meat paste can contribute to dimensional deviation of the printed structure and may affect the printing accuracy of tall designs. When extruding the material with a predetermined nozzle diameter, the springiness may affect the actual deposited stream diameter and hence vary the actual nozzle height, as the deposited object height significantly increases in comparison to the initial design.

Likewise, dragging, under- and over- deposition on the meat paste stream may be observed if the nozzle speed and extrusion rate are not properly set. The nozzle speed determines the movement rate of the print head, and needs to be adjusted with preliminary trials or by calculating the optimal nozzle speed (Hao et al., 2010). The extrusion rate (flow) determines the volume of deposited material per unit time (Wang & Shaw, 2005). At an optimal nozzle speed, the stream diameter equals that of the nozzle (Khalil & Sun, 2007). If the nozzle speed is too high, a thinner stream of meat paste is obtained and dragged, preventing the subsequent binding of layers and producing inaccuracies in the final product since voids remain within the cross-section area, and under deposition may occur. In addition, if the nozzle speed is too

low at a given extrusion rate, thicker streams are extruded and over deposition may be observed (Figure 6B). Furthermore, an increase extrusion rate produces more dense products due to higher amount of deposited materials and thus reduces the void fraction.

Similarly, varying infill percentages will affect the total amount of deposited material in the internal part of the printed structure, affecting the void fraction within the final 3D printed meat product and thus the post-processing conditions. For instance, the void fraction would determine the cooking conditions for a specific degree of doneness since as more porosity remains within the structure, less heat transfer occurs during cooking, affecting the moisture and fat releases and thus the texture of the cooked meat product.

In addition, the setting of the infill pattern actuate the stability of the printed meat product. For instance, a rectilinear or honeycomb pattern will provide more anchor points within the structure to allow the meat paste deposited as infill to bind with the vertical shell or perimeter due to the particular stickiness of the meat paste. On the contrary, unless an 80-100% infill percentage is used, the concentric pattern lack of anchor points between the infill material and the vertical shell in which the meat paste can bind to provide stability to the structure.

In general, while adjusting the above reviewed parameters essential for geometrical accuracy during 3D printing of meat, the economical aspect should also be considered. For instance, lower printing speed and nozzle diameter, as well as increased infill percentage, may result in higher accuracy, but longer printing times and energy consumption.

2.5 Design development

The in-software design for a determined 3D printed meat product sculpts its nutritional and sensorial profile. Through the combination of countless types of cross-sectional patterns, varying formulations and the combination of meat paste with different feed materials, 3D printing allows the development of new textures, mouthfeels, and nutritional composition (Sun et al., 2018) of meat products. According to Lipton et al. (2015), the texture of a food product can be modified by either combining materials with different textures into one pattern or by varying the added porosity of a printed meso-structure; while the nutritional composition can be customized based on data-driven recipes. Even though the rheology of the meat paste may represent a challenge when reproducing such complex patterns, these approaches could provide food consumers with both on-demand nutrition and novel eating experiences.

For instance, meat paste can be printed with a multi-head printer to include different ingredients in target locations/layers within the printed paste (Figure 7), such as salt, garlic, fatty slurries, etc. to contribute diverse mouthfeels and flavours.

Likewise, diverse food designs can provide those with chewing and swallowing difficulties with alternatives for the “ice cream-scooped” pureed food commonly served in age care facilities and hospitals (Cichero, 2015), being able to receive instead a meat product with modified texture suitable for their individual needs, with an appetizing appearance that resembles that of the original product. As an example, three hypothetical designs (Autodesk, Inc.), such as sausage, steak and beef patty are shown in Figure 8. In this way, recombined meats, such as steaks can be 3D printed as a multi-material model from soft meat paste, fat slurry and other food ingredients to approximate the flavours and nutrients of a beefsteak.

In order to do so, the expected final product have to be schemed as a CAD model first, and then converted into .STL format by a slicing software to be recognised by 3D printers. The model is sliced into 2D cross-sectional layers, according to the required design and printing settings (Noorani, 2017). Part of the layering sequence of a multi-material model (steak) is presented in Figure 9 (Repetier-Host V2.1.2 and Slic3r), where the yellow filament represents the meat paste and the turquoise filaments represent the fat. It is important to note that both, the meat paste and the fat portions intended to form the final construct must be schemed as independent CAD models, sharing the same coordinates, and then grouped in the slicing software as a single multi-material file, as suggested by Liu, Zhang, and Yang (2018b).

2.6 3D Printing methodologies suitable for meat materials

A variety of 3DP methods has been used for food printing, such as extrusion, inkjet printing, binding deposition, and bioprinting (Godoi et al., 2016), which are commonly applied to paste-like materials, liquid-based foods, powder-based foods, and cultured cells, respectively.

2.6.1 Extrusion

3DP by extrusion is the most applicable technology for food materials, which consists of ejecting the material through the nozzle in a digitally controlled way. The stream is deposited through cross-sectional stacking layers according to a previously designed pattern, until a 3D solid structure is obtained (Sun et al., 2018).

3D printing of meat products consists of building the desired geometry from a slurry material, which requires controlled temperature below 4 °C, calling for liquid-based methodologies, such as extrusion and/or inkjet printing. However, due to the limitation of inkjet printing for

the construction of complex structures (Pallottino, Hakola, Costa, Antonucci, Figorilli, Seisto, & Menesatti, 2016), extrusion is suggested as the most suitable methodology for 3D printing fibrous meat materials (Liu et al., 2018a). Among the available extrusion mechanisms (syringe-based, air pressure-driven and screw-based extrusion), air pressure driven extrusion is not recommended for viscous paste materials due to their ease of attaching to the walls of the cartridge (Sun et al., 2018), and thus is not endorsed for 3D printing of meat paste. The screw-based extrusion includes a screw conveyor that allows continuous feeding, mixing and deposition of meat material, while keeping the air entrapment to a minimal level (Sun et al., 2018). However, in order to make it suitable for highly viscous materials, such as meat paste, the screw blades should function alongside the conveyor unit and hopper to aid material displacement and avoid it from sticking to the hopper walls. Also, large and continuous amount of feeding material is needed in order to facilitate its displacement. On the other hand, in syringe-based extrusion, entrapped air within the meat paste and the cartridge can cause an increased compressibility during printing, resulting in inconsistent flow and inaccuracies (Lipton et al., 2010), although the air entrapment can be avoided in the first place during cartridge filling. In addition, a limitation of this mechanism is the inconvenience of replacing or refilling the cartridges, which prevents a continuous feed and limits the amount of material to be deposited per run, thus compromising the size of the printed meat product. Therefore, considering the suggestions mentioned above, both screw- and syringe-based extrusion seem more appropriate for 3D printing of meat.

2.6.2 Bio-printing

Bio-printing is a novel technology based on tissue engineering, still under development for food applications. Modern Meadow aims to obtain raw meat tissue by printing cultured stem cells. In this approach, an inkjet printer deposits cells into an agarose gel support structure, to be fused and form engineered meat (Forgacs et al., 2014). After fusion, the agarose structure is removed and the tissue is subjected to low-frequency stimulation in a bioreactor to mature meat fibres (Sher et al., 2015). Although, this methodology represents a great advancement to reduce slaughtering, it still needs to overcome challenges in matter of cost-effectiveness, sensorial attributes of the final products and consumers' acceptance.

3. Formulating the meat for enhancing its printability

3DP by extrusion requires non-Newtonian fluids showing shear-thinning behaviour in order to retain the desired printed shape (Lipton, 2017). The extrusion of meat paste involves the application of mechanical and shear stress in order to produce a strain that allows the

transport of the material along the hopper and through the nozzle (Yang et al., 2018).

Therefore, the assessment of the meat paste's rheological and mechanical properties helps to predict its behaviour during printing and deposition, and aids the setting of printing parameters. The printability of any food material refers to its ability to be handled and dispensed by a 3D printer as a freeform structure (Godoi et al., 2016). Printability is affected by several factors including temperature, printing parameters, and the rheological properties of food materials (Kim et al., 2017), making it challenging to describe the printability of a particular ink under varying printing conditions.

Although several studies reveal the assessment of different methodologies and foodstuffs in order to 3D print food products with diverse properties, one of the biggest challenges is the development of printable food formulations. A fine combination of an appropriate food formulation with suitable processing parameters must hinder the printability and structure stability issues.

Meat ingredients, which fit in the category of non-native printable traditional food materials, require a modification into a meat paste with suitable viscosity in order to be extruded from the nozzle and still be able to hold its structure upon deposition. Therefore, the expected printability, stability, and post-processing conditions need to be taken into account when designing the formulation.

First, as a fibrous material, the raw meat needs to be finely comminute into a paste form with controlled particle size to enable the extrusion through the nozzle of mm to micron size. The degree of comminution will depend on the type of the product to be printed and its textural characteristics. Meat mincing aids the extractions of myofibrillar proteins that assists the formation and stability of the batter emulsion through its interaction with other emulsion constituents. When working with off-cuts as raw material, the amount and particle size of connective tissue, as well as other non-meat tissues have to be considered, since it may affect the printability of the paste. In general, the particle size of the paste ingredients needs to be lower than the intended nozzle diameter for the printer to avoid clogging.

Additives, such as plasticizers may be required for the meat paste to be easily extruded, as well as binding components to adhere the subsequent layers once deposited. For instance, gelatine was added to a chicken, pork and fish slurry to enhance its printability (Liu et al., 2018a). Likewise, slow cooking and frying of 3D printed turkey and scallop pastes into

complex structures was feasible with the addition of TGase right before extrusion, acting as a heat-stable cold-set binder (Lipton et al., 2010).

3.1 Potential viscosity enhancers and binders for printable meat paste

The viscosity of the paste has to be low enough to flow easily through the nozzle and high enough to maintain the deposited shape (Godoi et al., 2016), and further support the subsequent layers on top. In order to control the viscosity of the paste, flow enhancers such as hydrocolloids and fats can be added. The use of hydrocolloids, including polysaccharides and proteins from plant, animal and microbial sources, is widely known in the meat processing industry where they act as thickeners, gelling and binding agents, syneresis controllers, emulsifiers, textures stabilizers, etc. (McArdle, Hamill, & Kerry, 2011). However, to improve the mechanical stability of the paste upon deposition, heat- and cold- set binders are available based on the temperature required for the occurrence of the binding mechanisms that are described below.

3.1.1 Cold-set meat binders

The use of cold-set binders allows producers to lower the amounts of salt components (salt/phosphate technology) in meat product formulations and restructure lower-value pieces of muscle into value-added products, such as “restructured meats”.

For the addition of cold-set binders into the meat batter, the formation of a heat-resistant gel may be required, depending on the intended post-processing conditions of the 3D construct. If a thermo-reversible gel is obtained, the structure of the construct could be lost. However, the addition of thermo-reversible gelling agents may be overcome with the combination of irreversible-gel producer heat-induced binders. Some cold binding systems producing heat-resistant gels include the enzyme system (TGase), calcium/alginate system, and plasma protein system (fibrinogen/thrombin).

TGase is an enzyme obtained from animal blood or bacterial fermentation which catalyses cross-linking of proteins and peptides, and thus is commonly known as meat glue (Boles, 2011). Microbial TGase (mTGase) is enzyme system applicable for meat cold binding mechanisms based on the covalent cross-linking of glutamine and lysine in protein molecules (Payne, 2009), resulting in the modification of physical and chemical properties of food products, such as viscosity, firmness, thermal stability, elasticity and water holding capacity. Although the enzyme has been isolated from different sources, the microbial-based represents more profitability for the industry and thus is more widely used (Boles, 2011). mTGase has

an activity temperature ranging from 0 to 60°C, and pH activity range of 4.5-8.0, with favourable conditions being 40°C at pH 5.5, and isoelectric point of 8.9 (Kieliszek & Misiewicz, 2014). The binding mechanism is enhanced by the presence of sodium caseinate, gelatine, soy proteins, myosin, salt and phosphates, and depending on the dose of usage, the substrate and the binding conditions, it may take up to 24 hours to obtain the desired final state (Payne, 2009). Since the cross-linking process can start within 20 min (according to the doses) of contact, adding TGase to the mixture right before extrusion prevents the paste from binding before deposition has occurred. Furthermore, the addition of salts and phosphates is recommended to aid the extraction of salt soluble proteins, such as myofibrillar and some sarcoplasmic (Boles, 2011), and thus increase the binding matrix. For instance, the addition of NaCl resulted in reduced gel setting time (from 1.5 to 1 h) for porcine meat batters containing 1% mTGase with increased water binding capacity (Sadeghi-Mehr, Raudsepp, Brüggemann, Lautenschlaeger, & Drusch, 2018).

The presence of constituents with water holding capacity, such as gelatin, may enhance the viscosity of the paste to make it extrudable, while the addition of TGase shortly before printing would help retain the shape of the 3D construct during deposition and post-processing. Although the moisture release upon gelatin melting may represent a challenge.

On the other hand, the calcium/alginate system is composed of sodium alginate, a source of calcium (e.g. calcium carbonate) and an acidifier (e.g. lactic acid) to aid the slow-release of calcium. When in contact with meat products under moulded shaping, it slowly forms an irreversible gel taking from 2 to 48 hours at 0-5°C for the gel to set, depending on the solubility of the calcium source (Boles, 2011; Means & Schmidt, 1986). The recommended concentration for structured meats is 1.5%, allowing the addition of 1% salt and 35% water (BDF Ingredients, n.d.).

Fibrinogen/thrombin (FT) (10:1 or 20:1) is a plasma protein system derived from the blood clotting mechanism in which fibrinogen is enzymatically converted to fibrin by thrombin. The two components in the system are packed and frozen individually and require thawing until a temperature of 26.6 °C is reached before contact with the meat pieces. The mixture is rapidly moulded since it takes around 10 to 15 minutes before reaction starts, requiring a minimum of 5 hours for maximum gel strength with a typical usage dose of 5-10% depending on the dimensions of the meat pieces (Boles, 2011). In the case of meat emulsions, the contact time for maximum strength may vary depending on the FT doses and the interaction

with other components in the matrix. For instance, in pork meat batter added with 10-20% FT, increased hardness was observed after 12h of storage at 6 °C for mixtures with 0.25-0.5% NaCl compared to those with 2% NaCl. Such results were suggested to be attributable to the partial solubilisation of myofibrillar proteins without affecting the fibrin gel formation since further addition of NaCl reduced the breaking strength values of the fibrin gels (Romero de Ávila, Hoz, Ordóñez, & Cambero, 2014).

Cold-swelling hydrocolloids, such as xanthan gum, guar gum and gum tragacanth may act as viscosity and extrusion enhancers through the stabilization, thickening and emulsification of the paste, however, they do not act as gelling agents independently (Feiner, 2006). If employing hydrocolloids blends to attain a gel, the gel heat-resistance must be studied to ensure that the 3D construct is not damaged during cooking. Some hydrocolloid blending applied to meat products include basil seed gum/gelatin (Lee & Chin, 2017), carboxymethylcellulose/locust beam gum/ xanthan gum (Chattong, Apichartsrangkoon, Chaikham, Supavititpatana, & Bell, 2015), konjac glucommanan/guar gum (Zhao, Yang, Sun, & Cong, 2014), and carrageenan/locust beam gum (Zhuang, Yang, Sun, & Chen, 2013), among others.

3.1.2 Heat-set meat binders

Restructured meat products are traditionally dependant on the extraction of myofibrillar proteins by means of ionic strength through the addition of salts, followed by thermal gelation to maintain the moulded shape of the product (Boles, 2011). However, other technologies rely on the thermal gelation or coagulation of the entire system by the addition of heat-set binders, such as blood proteins and other hot-swelling hydrocolloids. Although other hydrocolloid sources including milk (Haast, Morressey, & Fox, 1987), eggs (Li, Li, Wang, Zhang, Xu, Zhou, Su, & Yang, 2017) and soy proteins (Maltais, Remondetto, Gonzalez, & Subirade, 2005) are available, their allergenic issue needs to be considered.

Blood plasma proteins (BPP) represent 6-8% of plasma weight and are used in meat products as binding agents for having gelation, emulsification and solubility properties, which can be mainly accredited to its albumin (up to 60%), globulin (40%) and fibrinogen (around 3%) content (Parés, Saguer, & Carretero, 2011; Tarté, 2009). BPP are highly soluble at pH values above 5.5-6.0 (Zayas, 1997) and its pH value of 7.4-7.8 raises the pH value of the meat product to some extent, thus increasing its water binding capacity. In meat products, concentrations of 0.5-2% (w/w) of BPP provide a heat-stable gel, starting at 64°C and

reaching its maximum strength at 72°C (Feiner, 2006). Although further increase of temperature (>95 °C) may harden the gel, meat products are not exposed to temperatures beyond 70-75°C (Tarté, 2009). At such conditions, the presence of BPP within the printable paste would aid in the formation of an irreversible gel, maintaining the 3D printing shape during post-processing.

Consequently, in order to modify the rheological and mechanical properties of the meat paste for 3DP, the emulsifying and gelling properties are of primary importance, respectively. Cold-set binders that provide heat-resistant gels may be used to attain the modification of both, rheological and mechanical properties. While, heat-set binders can be added to the paste to enhance its mechanical properties mostly during post-deposition and post-processing operations.

Other hydrocolloids such as polysaccharide gums modify the viscosity of the paste through water retention and gel formation. Such ability to retain water serves as a lubricant that simulates the sensorial perception of fat and thus polysaccharide gums are known as fat replacers in low-fat processed meats (McArdle et al., 2011), without interfering with protein activation in meat products (Feiner, 2006). For instance, carrageenan, mainly kappa and iota types and their salts, are widely used as binders in low-fat meat products for their gelling, thickening and stabilizing properties during the heating stage, while lambda type is considered a non-gelling agent (Paglarini, Furtado, Biachi, Vidal, Martini, Forte, Cunha, & Pollonio, 2018; Trius, Sebranek, & Lanier, 1996). However, for the post processing operations of 3D printed meat products, the physical properties of hydrocolloids such as solubility/swelling/melting and gel properties are of utmost importance and their contribution to the rheology of the meat paste formulation must be extensively studied. In addition, the synergism observed in heat-swelling hydrocolloids blends observed in water gel systems may not be effective for meat systems. A hydrocolloid swelling temperature higher than that of the proteins coagulation temperature, may prevent the even distribution of the hydrocolloid within the matrix, and its interaction with other constituents, as reported by Prabhu and Sebranek (1997) when studying the effect of carrageenan and starch on turkey ham product.

In general, the addition of different food hydrocolloids to the meat paste can provide modified rheological and mechanical properties through varying binding mechanisms, enhancing its printability and post-processing viability.

4. Conclusion

Although 3DP technology has been widely applied for several types of food materials, very few studies refer to the printability of fibrous meat materials, such as pork, turkey, chicken and fish, while no data is available for beef meat. Information regarding the formulation to adjust the rheological and mechanical properties for beef paste is required for a better understanding of its printability, as well as the 3DP settings and post-processing conditions of the printed product. With the existence of such comprehensive data, further research may be conducted with beef materials in order to improve its nutritional value and sensorial profile by means of addition of bioactive ingredients and including complex internal structures, respectively. Furthermore, an adequate 3D printer for meat products is suggested to be extruder-type consisting of screw conveyor or syringe system with uninterrupted temperature control throughout the feeding system, the hopper, the nozzle and the platform itself in order to reduce food safety risks and controlling the material's rheology during the printing process.

5. Competing interest statement

None to declare

6. Declarations of interest

None

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Figure 1 Categories of food printability (Sun et al., 2015)

Figure 2 Turkey with TGase was printed into a (a) truncated hemisphere and (b) cooked sous-vide. Celery fluid gel (green fluid in c) was printed within the structure of a (d) turkey cube (Lipton et al., 2010). Image reproduced with permission from David Bourell, <http://sffsymposium.engr.utexas.edu/>

Figure 3 Fibrous materials printed into block shape (ball screw-based printer) (Liu et al., 2018a). Image used under the terms of the Creative Commons Attribution 3.0 licence

Figure 4 3D printed surimi gel with different NaCl concentrations (A= 0%, B= 0.5%, C= 1%, D= 1.5%). Printing settings: 2.0 mm nozzle diameter, 5.0 mm layer height, 28 mm/s moving speed and 0.003cm³/s extrusion rate. Reproduced from Wang et al. (2018) with permission from Elsevier

Figure 5 Extruder type - 3D printing stage with Cartesian configuration: (a) single nozzle type, (b) dual-nozzle type

Figure 6 Streamline accuracy with A: varying nozzle heights and B: varying printing speeds: Scenario 1 ($V1 < VN$), Scenario N (VN), and Scenario 2 ($V2 < VN$). Adapted from (Khalil & Sun, 2007; Severini et al., 2016a)

Figure 7 Multi-material CAD model

Figure 8 Hypothetical food designs for age care homes: (a) sausage, (b) steak 'recombined meat', and (c) patty

Figure 9 Cross-sectional layers for multi-material model (steak)

Highlights

- Multi-material 3D printing allows the production of recombined meats.
- The design of appetizing soft-meat products is viable with 3D printing technology.
- Low temperature-3D printers are needed to process meat products safely.
- The application of heat- and cold-set binders enhances the meat paste rheology.

ACCEPTED MANUSCRIPT

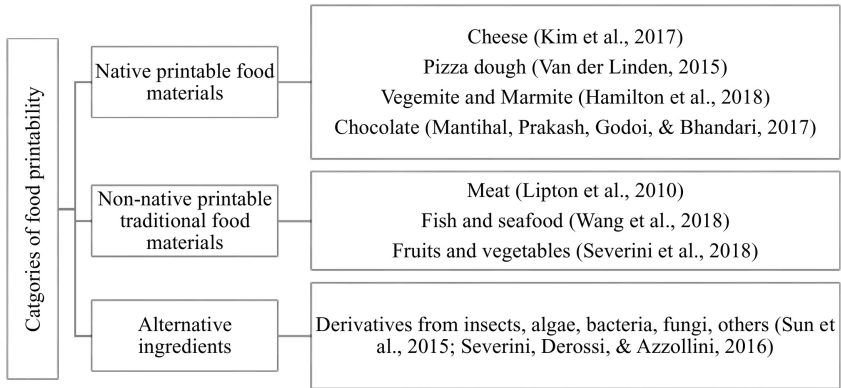


Figure 1

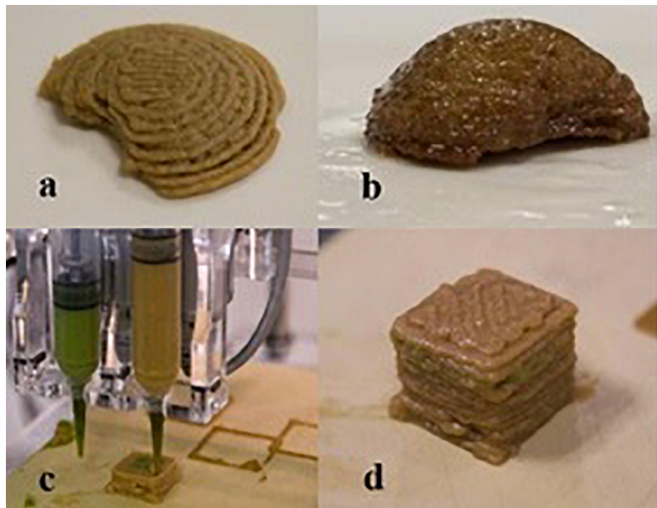


Figure 2



Figure 3

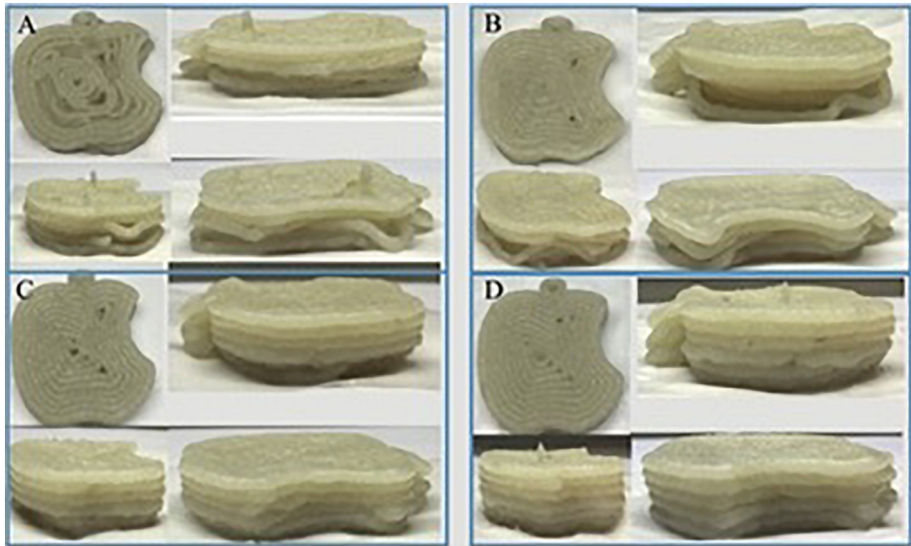


Figure 4

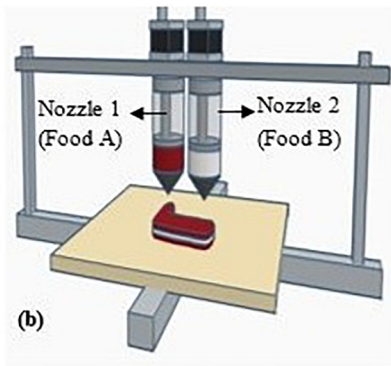
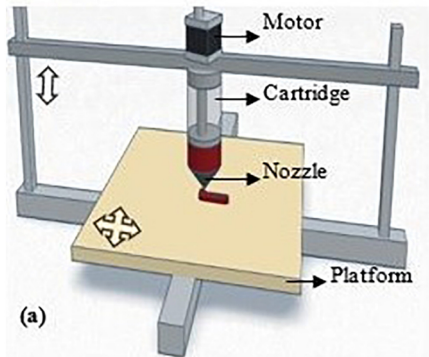
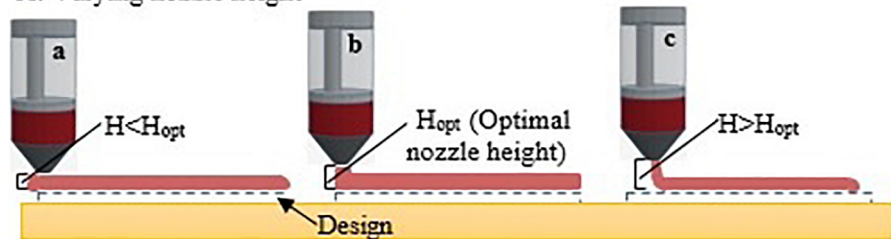


Figure 5

A: Varying nozzle height



B: Varying printing speed

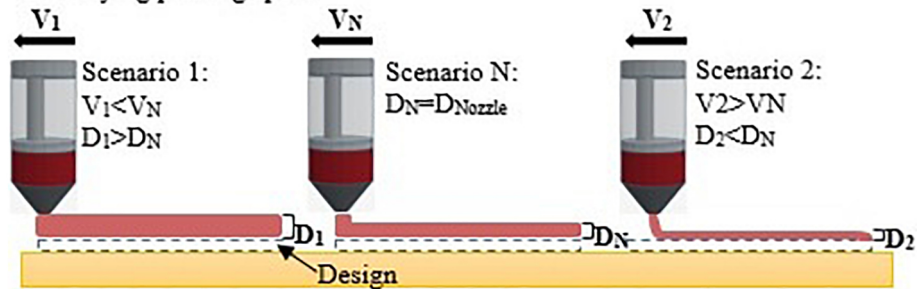


Figure 6

Salt slurry

Garlic slurry



Meat paste

Fat slurry / cheese

Figure 7

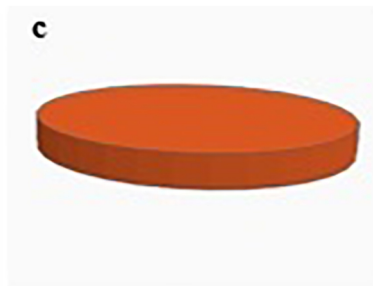
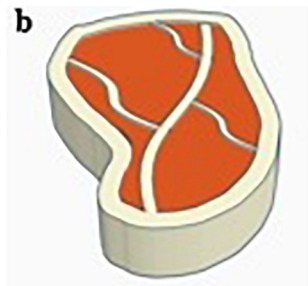
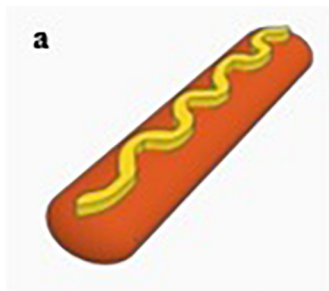


Figure 8

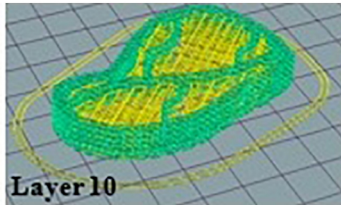
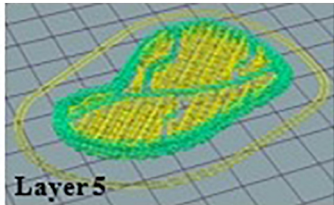
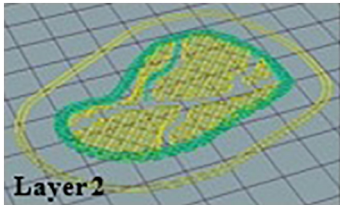


Figure 9