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Review of 3D printing and potential red meat applications

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Abstract

Project V.RMH.0034 reviews the fundamentals of 3D printing (3DP) technology, meat science and conventional meat by-products processing. The knowledge acquired has enabled us to consider the extrusion 3D deposition the most suitable method to be used on red meat. The build material can be prepared from meat protein mixture (wholesome, offcuts and offal) in combination with hydrocolloids from different sources (e.g. animal, dairy, plants, alginates) or slurry-mixed with the meat powder developed by MLA (projects A.MPT.0036 and A.MPT.0060). By the end of the project, a value proposition was structured aiming to achieve efficient utilization of meat by-products by integrating 3DP with existing technologies for meat by-products utilization. The mentioned research proposal will give added value to the meat supply chain by: (i) Introducing in the food service sector an innovative edible product based on meat ingredients; (ii) Providing applicability to processed meat derivatives and (iii) Using meat by-products to manufacture healthy food and new textures. The main advantage of 3DP technology can be related to the preservation of meat protein and micronutrient profile during processing; enabling the manufacture of healthy food rich in iron, zinc and proteins. Project V.RMH.0034 is expected to suggest MLA to support future investment decisions and undertake ground breaking research on forefront 3DP technology using meat and its by-product as ingredients.

Executive Summary

The application of novel and smart manufacturing technologies using meat and its by-products are needed. This strategy can increase competiveness by lowering production costs and improving sustainability throughout the red meat supply chain. Based on that, project V.RMH.0034 was undertaken to provide knowledge on the breakthrough 3D printing (3DP) technology and propose adaptations for the application of meat ingredients as material supply. The specific objectives involved in this study can be listed as follows:

- Describe science fundamentals of 3DP works in both food and packaging applications;
- Complete brief literature review and patent search to identify emerging areas for this platform;
- Present overview of global applications and network of providers in Australia;
- Describe meat science fundamentals to enable 3DP;
- Define the Value Proposition for red meat and 3DP and recommended next steps.

3DP technology

3DP technology, a popular term used for additive manufacturing (AM), is defined by the process of fabricating a physical object from a three-dimensional digital model, by depositing consecutive thin layers of a material. Depending on the state of the material supply (liquid or powder) and the required end-use properties, a number of 3DP techniques can be applied to achieve self-supporting layers upon deposition and post-processing. In the past few years 3DP technology has gained particular attention in the aerospace and biomedical fields. In the food sector, 3DP is considered an emerging technology which gives freedom to design innovative edible product in a variety of shapes, flavours, nutritional content and texture. Our patent search revealed 81 patents describing methods to print 3D structures using food. With 64 patents, the liquid-based techniques represent the majority of the AM processes.

3DP technology has also brought innovation to the packaging industry. Our brief literature review showed that 3DP technology has been used for prototyping blowing molds, fibre filling molds and thermoforming tools. In the field of intelligent packaging, a smart cap, fabricated using 3DP technology, has enabled to detect when milk deteriorates. As an example of active packaging, we have also observed that 3DP technology is receiving much attention to fabricate microwavable packaging using metal-based conductive inks.

Meat science and existing meat by-products technologies to enable 3DP

The identification of possible applications of 3DP technology on red meat was based on two key factors:

(1) **Meat science fundamentals**: The research team has reviewed the functional properties of the main constituents of meat (proteins and fat). This knowledge was pursued to help understanding the binding mechanisms involved between layering deposition when meat ingredients are used as supply material.

(2) **Review of existing meat by-products technologies**: Available information about the existing meat by-products processing were collected and related with the functional properties of meat constituents. This approach aggregated a strong

scientific background which enabled the composition of a value proposition for MLA stakeholders.

Value proposition for MLA stakeholders

The value proposition suggests the use of raw-meat, meat off-cuts and powdered meat from projects A.MPT.0036 and A.MPT.0060 as raw-materials to prepare printable "inks". For a profitable use of powdered meat, it is envisaged future collaboration with companies that have already contributed for the fabrication of powdered meat together with MLA, such as, Mahltechnik Gorgens GmbH (Germany) and Thricor Pty Ltd (Australia).

The value proposition aims to achieve efficient utilization of meat by-products by integrating 3DP with existing reforming meat technologies (**Fig. 1**). This research will give added value to the meat supply chain by:

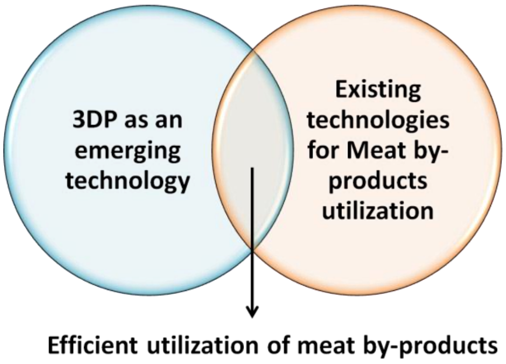

Figure 1: 3DP in an integrated platform with existing technologies of meat by-products utilization.

- Introducing in the food service sector an innovative edible product based on meat ingredients;
- Providing applicability to processed meat derivatives, such as, powdered meat, gelatin, collagen hydrolysates and blood plasma proteins;
- Using meat by-products to manufacture healthy food and new textures;

Penetration levels of the designed materials in different groups of the food service sector (take-away, dining out, event/leisure and institutional) might determine the required end-properties of the edible 3D construct. Particular attention will be devoted to create new textures for aged care homes; due to the difficulty faced by elderly people to chew and swallow food.

Future work

Project V.RMH.0034 will help MLA to support future investment decisions and undertake ground breaking research about 3DP technology using meat as ingredient. 3DP technology was recognized as a powerful approach to add value to the meat supply chain because of meat by-products can be used to create new textures and flavours. It is therefore recommended to perform further R&D work to establish the development of a cost-effective and scalable manufacturing process based on the efficient utilization of meat by-products. Future work will involve the optimization of the build material using the following meat by-products in a slurry consistency for application in 3DP extrusion-based processes:

- **Raw meat and off-cuts**: The raw meat and/or off-cuts will be used to make the printable paste. Combinations of appropriate ingredients listed below will be added considering the nutritional composition, self-supporting structure after deposition and textural attributes of the final product.
- **Powdered meat**: previous studies have shown the high protein, zinc and iron content of powdered meat. By using 3DP technology powdered meat can be

converted into a high-value aggregated product without losing its protein and micronutrients profile (A.MPT.0036 and A.MPT.0060);

- **Collagen derivatives**: chemically and enzimatically hydrolysed collagen can be used to control the viscosity of the printable material. Particular attention will be devoted to enzymatically hydrolysed collagen (ECH) which is soluble at ambient temperature, free-from chemical residues and completely digested in the small intestine. The use of ECH will afford a healthy characteristic to the end-product as it is known that the consumption ECH can prevent muscular loss. The molecular mass of ECH will be varied by conducting different levels of hydrolysis which will result in variations on the viscosity of the ECH solution (A.BIT.0011).
- **Blood plasma protein (BPPs)**: liquid (P.PSH.0415) and powder (A.BIT.0016 and A.BIT.00200) blood plasma proteins can be used to enhance the rheological properties of the formulation to be extruded. Previous work have highlighted the ability of BPPs of binding meat pieces in conventional restructured meat processes (A.BIT.0015 and A.BIT.0020). By analogy, this property has potential to enhance the interactions between layers, providing an stable 3D construct;
- **Tallow and lard**: The fat content of the 3D printed meat-based product will be controlled by varying the ratio between rendered lard (low melting point) and tallow (high melting point). “Healthier” tallow which consists of unsaturated fatty fraction of tallow obtained from fractionation or transesterification processes can also be used to incorporate flavour without increasing the content of unsaturated fatty acids (A.COP.0067).

By using the ingredients described above, 3DP will be automatically integrated into a platform of existing technologies to process meat by-products. The findings from future work will assist the red meat processing sector in enhancing their sustainability and profitability (i.e., through capturing more value from under-utilised co-products). Gaining early competence in the field of 3DP technology is essential to MLA play a decisive role in the new generation of scientific knowledge and fully capitalise upon the development of new technologies.

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1 Background

Three dimensional printing (3DP) was originally invented to build 3D objects based on materials, such as, metals, ceramics and polymers aiming to perform the fabrication of complexes parts in a single step. In the very first studies, the fabrication of 3D objects from polymers relied on photo-polymerization processes in which ultraviolet curable polymers were used for printing layers upon layers of solid constructs (Kodama, 1981, Hull, 1986). In the past few years 3DP technology has gained particular attention in the aerospace and biomedical fields. The benefit of using this technology relies on the free-form design of complex and lighter parts which could not be fabricated through conventional manufacturing processes. In the biomedical area, thousands of people are being benefited by receiving 3D printed implants using biocompatible materials such as titanium and ceramics. The main advantage can be related to the fact that implants are designed according to the specific characteristics of each human body parts.

Our recent review on 3DP technology to design food shows that the food industry will be gradually incorporating 3DP as a breakthrough technology. The curiosity from various multinational companies, research institutions and universities is growing rapidly. Numerous types of 3D printer fabrications for food application are being reported. 3D food shapes have been successfully printed by using liquid (e.g., melted chocolate, dough and meat puree) and powder (e.g., sugar and chocolate, as fine particles) based suppliers. However, the production of 3D printed food constructs is still challenging due to complexity shown by the multicomponent food systems. Aiming to understand the mechanisms involved in the formation of self-supporting layer throughout the 3D printing of a food material, in our review (Godoi et al., 2015) we tried to relate the principles of the technology with the properties of the essential food constituents (carbohydrates, proteins and fat). We suggested that the rational design of 3D food constructs relies on three key factors: (1) printability, (2) applicability and (3) post-processing (Godoi et al., 2015).

Project V.MRH.0034 comprised of a collaborative research focused on bridging the industry need of finding application for meat by-products with academic capabilities by establishing knowledge transfer networks. The transition of building meat-based materials onto a platform of solutions will be made possible by understanding the behavior of meat constituents on 3DP technology and conventional restructured meat processes. This final report presents an overview of the preceding milestone outcomes, define a Value Proposition for 3DP applied on red meat and recommended next steps for future work.

2 Project Objectives

(1) Describe science fundamentals on how 3D printing works in both food and packaging applications

(2) Complete brief literature review and patent search to identify emerging areas for this platform

(3) Present overview of global applications and network of providers in Australia

(4) Describe meat science fundamentals to enable 3D printing – consider water activity, pH, lean meat: fat, grissel, texture, juiciness, flavour etc and review combination

technology platforms such as Powdered Meat, HMEC, Tallow extraction, Emulsified/Textured Modified Foods as potential components - describe learnings, barriers and gaps and opportunities

(5) Define the Value Proposition for red meat and 3DP and recommended next steps.

3 Methodology

3.1 Literature review

The research team conducted a literature review to ascertain the current state of 3DP technology from an empirical and theoretical perspectives. Key topics included:

- Current 3DP technologies and materials supplies;
- Emerging 3DP applications;
- 3DP applicability in both food and packaging;
- Meat science fundamentals and current processes of reforming meat.

The concepts reviewed were used to identify potential applicatility of 3DP technology on red meat

3.2 Patent search and list of providers and applications

As part of MS1, a patent search was conducted by consulting the following databases:

(1) Derwent innovations index: One of the world's most comprehensive databases of patents information. Over 11 million basic inventions and 20 million patents are covered from over 40 patent-issuing authorities dating back to 1963. Derwent provides access to the full text of most patents.

(2) Google Patents: Patents from the United States or the European patent offices.

A total of 81 patents describing methods of AM applied to print 3D structures of food were found. The patents are listed in two tables (**Appendix I**) according to the material supply: liquid or powder. With 64 patents, the liquid-based techniques represent the majority of the AM processes. **Appendix II** contains the list of providers and applications.

4 Fundamentals of 3DP technology

3D printing (3DP) technology, a popular term used for additive manufacturing (AM), is defined by the process of fabricating a physical object from a three-dimensional digital model, by depositing consecutive thin layers of a material. Depending on the state of the material supply (liquid or powder) and the required end-use properties, a number of 3DP techniques can be applied to achieve self-supporting layers upon deposition and post-processing. **Table 1** lists the current 3DP technologies and their feasibility to print edible materials. The 3DP techniques are grouped according to their material supply: (i) liquid, (ii) filament or soft materials, (iii) solid and (iv) cells.

3DP technologies relying on photo-polymerization processes in which ultraviolet curable polymers were used for printing layers upon layers of solid constructs (Kodama, 1981, Hull, 1986) are not suitable to design food. Therefore, Stereolithography (SLA) and Multi-Jet

Modeling (MJM) are examples of 3DP technologies not suitable to print edible materials. However, curable printing inks can be attractive in the field of food packaging where there is a continued need for safer, faster and cheaper inks, functional coating, and overprint varnishes.

4.1 Supply: liquid

SLA is defined by the conversion of a liquid photosensitive resin to a solid state by selective exposure of a resin vat to ultraviolet (UV) light. In this process, a CAD model is sliced into layers, each of which then is scanned by the UV light to cure the resin selectively for each cross-section. After a layer is built, the platform descends by one layer thickness. Then, a resinfilled blade sweeps across the part's cross-section, recoating it with one layer thickness of fresh resin. The subsequent layer then is scanned, adhering to the previous layer. Different from SLA, MJM uses a print head with multiple nozzles to generate jets oriented in a linear array. Each individual jet dispenses UV curable polymer (or wax) on demand. The MJM head shuttles back and forth to build each single layer, followed by a UV lamp flashing to cure the deposited polymer. When one layer is completed, the platform is descended by one layer thickness and the next layer is built upon the previous layer. This process is repeated until the entire part is built (Guo and Leu, 2013).

InkJet printing (IJP) relies on the fundamental of accumulation of droplets of material deposited on-demand by ink-jet printing nozzles (Kruth, 2007). Inkjet printers generally operate using thermal or piezoelectric heads. In a thermal inkjet printer, the print head is electrically heated to generate pulses of pressure that push droplets from the nozzle. Piezoelectric inkjet printers contain a piezoelectric crystal inside the print head which creates an acoustic wave to separate the liquid into droplets at even intervals. Employing a voltage to a piezoelectric material arouses a prompt change in shape, which in succession produces the pressure necessary to eject droplets from the nozzle (Murphy and Atala, 2014).

4.2 Supply: polymeric filament or soft materials

The application of extrusion processes into additive manufacturing was introduced by the Fused Deposition Modelling (FDM^{TM}) method developed by Crump (Crump, 1991, Crump, 1992) and trademarked by Stratasys Inc (Batchelder, 2012). In this method a moving nozzle is used to extrude a hot-melt filament polymer as a continuous melted threat, fusing it to the preceding layer on cooling. This 3DP technology is not applicable for edible materials.

Robocasting was introduced by Sandia National Laboratories to extrude aqueous ceramic pastes layer by layer giving rise to a 3D shape. Typically, ceramic paste is extruded through a nozzle and deposited on a substrate. After the deposition of each layer, the vertical axis of the gantry system moves up by one layer thickness, and the next layer is deposited. This step repeats until the complete part is built. Control of paste properties is essential for the robocasting process. The paste dries from a fluid-like state to a solid-like state normally within 10 to 15 s of being deposited so that the next layer can be added without a long wait. If the paste is too thin, the deposits will come out as liquid beads that spread uncontrollably. If it is too thick, the deposits will look like rope. With proper paste viscosity and consistency, each deposited layer maybe a rectangular cross-section with relatively straight walls and flat tops (Guo and Leu, 2013). This technology has been adapted to print food constructs by researchers from Cornell University who introduced the Fab@Home Model 1 as an open source design 3D printer capable of producing forms using a liquid food materials (Periard et al., 2007, Malone and Lipson, 2007). In subsequent years many studies were carried out in an effort to adapt AM technology to the design of food constructs (Schaal, 2007, Hao et al., 2010b, Hao et al., 2010a, Diaz et al., 2014b, Diaz et al., 2015a, Grood et al., 2013, Sol et al., 2015, Serizawa et al., 2014). This represents a challenge because 3DP is not easily applied to the complex food materials with a wide variation in physico-chemical properties.

When the binding mechanisms of the accumulated layers during robocasting involves gel-forming ingredients; robocasting can be called hydrogel-forming extrusion. Another possibility is melting the material supply before loading it into an extruder; by this way the formation of layered structure is governed by solidification by cooling. More details about hydrogel-forming and melting extrusion 3DP technologies are given in the **Section 6.1**.

4.3 Supply: solid

Selective Laser Sintering (SLS) applies a laser, as a power source, to sinter the powder in a bed. A solid structure is built by directing the laser at points pre-determined by a 3D model. The laser partially melts the particle and fuses together specific areas of the powder-bed by scanning cross-sections. Afterwards, the powder-bed is lowered by one layer thickness, a new layer of particles is deposited on top, and the process is repeated until the object is finalized. SLS can produce parts from a relatively wide range of powder materials, including wax, polymers, polymer/glass composites, polymer/metal powders, metals, and ceramics.

The interaction between the laser beam and the particles used in SLS is important to define the feasibility and quality of any SLS process (Kruth, 2007). The selection of the laser has a relevant influence on the fusion of powder for two main reasons: (1) the laser absorptivity of materials relies on the laser wavelength (2) the mechanism for powder densification is affected by the input laser energy density (Gu, 2012).

Unlike SLS, Electron Beam Melting (EBM) completely melts the particles together because it applies a higher power laser beam to fuse the particles together. EBM process uses an electron beam rather than a laser beam as its energy source. EBM builds parts by melting metal powder layer by layer with an electron beam in a high vacuum chamber. The fabricated parts are fully dense, free of voids, and extremely strong (Guo and Leu, 2013).

During the fabrication by Binder Jetting (BJ), a liquid binder is ejected by a drop-on-demand print head onto a thin layer of powder following a sliced 2D profile generated by a computer 3D model. The binder plays an important role of joining adjacent particles together creating, therefore, a 3D construct. This can occur due to the dissolution-fusion or cross-linking of the particle surfaces (Peltola et al., 2008). Both BJ and SLS techniques require an additional step for removing the unfused material at the end of construction. **Fig. 2** shows a schematic representation of SLS and BJ methods.

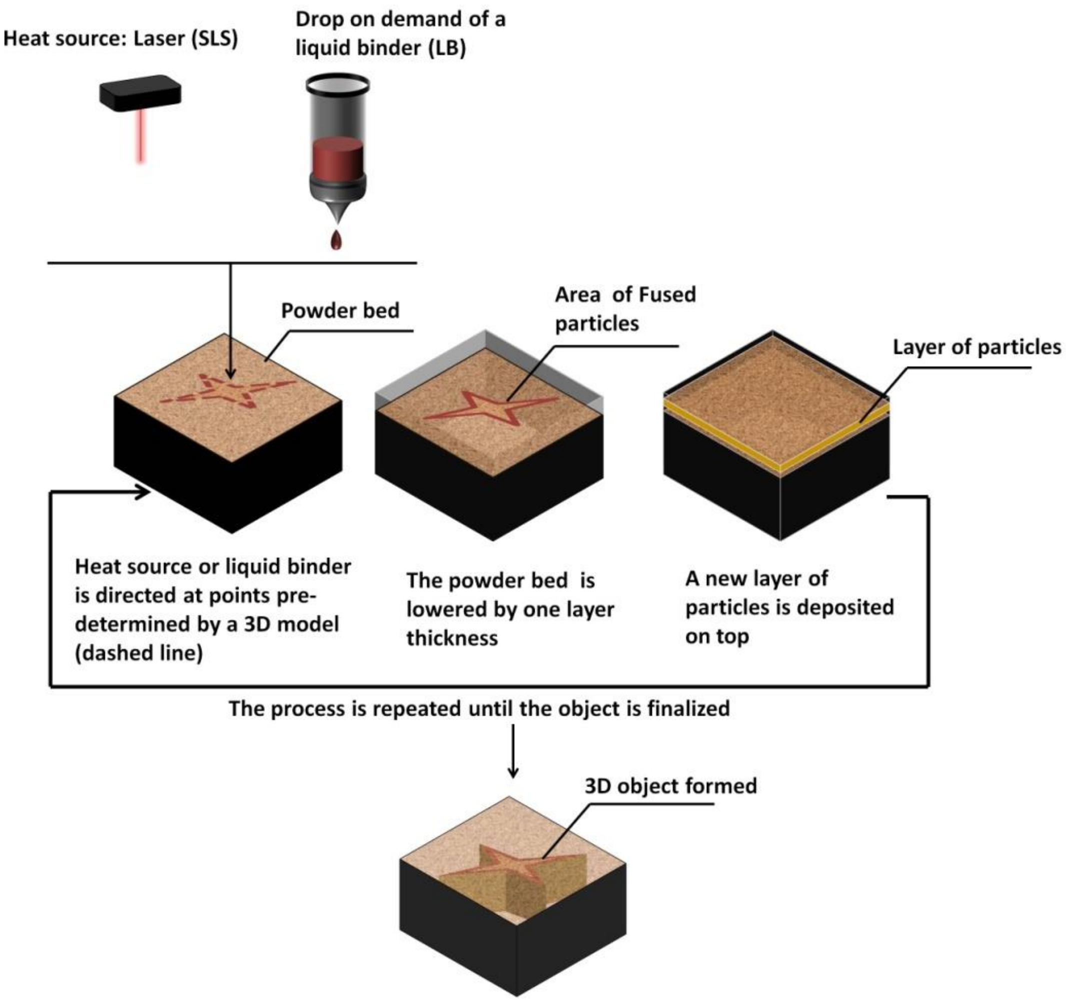


Figure 2: Powder-based 3D printing techniques: (a) SLS and (b) BJ.

4.4 Supply: cells

Bio-printing have been originally applied to build tissues without any biomaterial-based scaffold. This technique relies on the precise layer-by-layer deposition of biological materials and culture of living cells. The most common technologies used for deposition and patterning of biological materials are inkjet, microextrusion and laser-assisted printing (Murphy and Atala, 2014).

Considering meat as a post-mortem tissue, researchers from University of Missouri led by Prof Gabor Forgacs proposed to construct a strip of edible porcine tissue using 3D printing technology. Their technology uses multicellular cylinders as building blocks and thus depends on self-adhering cell types. Droplets of freshly prepared multicellular aggregates (the bio-ink particles) are deposited on-demand via an inkjet nozzle into a biocompatible support structure (in this case, agarose rods). The final construct is transferred to special purpose bioreactor for further maintenance and maturation to make it appropriate for use. During maturation, the bioreactor promotes pulsatile flow and the maturing graft develops biomechanical properties (**Fig. 3**) (Marga et al., 2012, Norotte et al., 2009, Forgacs et al., 2014).

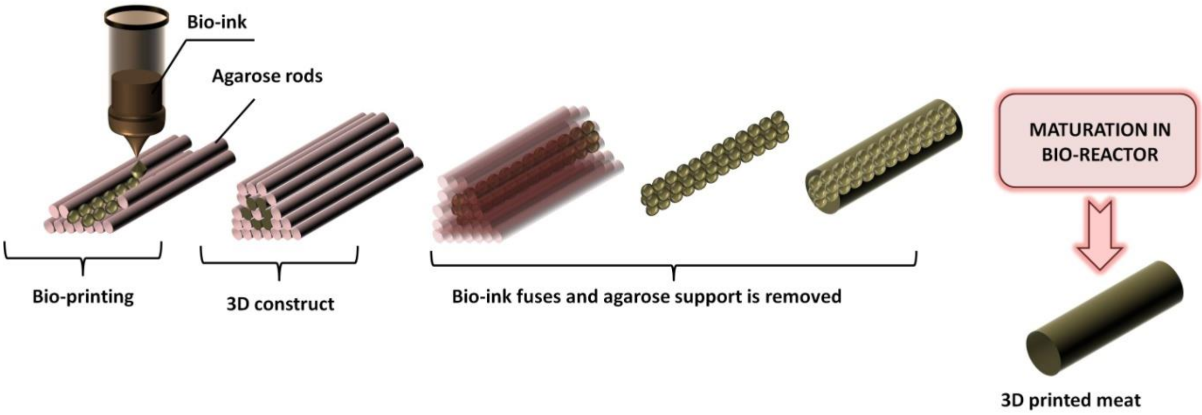


Figure 3: Schematic illustration of bio-printing technology applied to construct meat.

Upon affordable price, in the future this technology would benefit the masses with religious restrictions on meat consumption and populations with restrict access to safe meat production. The bio-printing of meat, however, shows many drawbacks to overcome, most of them associated to the spatial resolution of the final construct and long maturation processes (Marga, 2012).

Table 1: Current 3DP technologies and their feasibility to print edible materials. Adapted of (Guo and Leu, 2013).

Technique	Material preparation	Principle	Binding mechanisms	Typical materials	Edible materials
Supply: liquid					
Stereolithography (SLA)	Liquid resin in a vat	Laser scanning/light projection	Photopolymerization	UV curable resin, ceramic suspension	Not applicable
Multi-Jet Modelling (MJM)	Liquid polymer in jet	Ink-jet printing	Cooling and photopolymerization	UV curable acrylic plastic, wax	Not applicable
InkJet Printing (IJ)	Liquid droplet in nozzle	Drop-on-demand deposition	No phase change, generally used to print patterns or filling cavities	Hydrocolloids composite for medical applications, metal-based conductive inks	Chocolate, liquid dough, sugar icing, meat paste, cheese, jams, gels
Supply: polymeric filament/soft materials					
Fused Deposition Modelling (FDM)	Filament melted in nozzle	Extrusion and deposition	Solidification upon cooling	Thermoplastics, waxes	Not applicable
Robocasting	Sof-material in nozzle	Extrusion and deposition	No phase change, accommodation of layers relies on rheological properties	Ceramic paste	Frosting, processed cheese, dough, meat puree
Hydrogel-forming Extrusion (HFE)	Sof-material in nozzle	Extrusion and deposition	Ionic or enzymic cross-linking	Hydrocolloids composite for medical applications	Xanthan gum and gelatine
Melting Extrusion (ME)	Sof-material in nozzle	Extrusion and deposition	Solidification upon cooling	-	Chocolate
Supply: powder					
Selective Laser Sintering (SLS)	Powder in bed	Laser scanning	Partial melting	Metal, polymers	Sugar, Nesquik
Electron beam melting (EBM)	Powder in bed	Electron beam scanning;	Full melting	Metal	Not applicable
Binder Jetting (BJ)	Powder in bed	Drop-on-demand binder printing	Adhesive forces or chemical reactions between powder and binder	Polymer, Metal, ceramic, other powders	
Supply: cells					
Bioprinting	Cell culture	Drop-on-demand deposition	Self-assembly of the cells	Cells	-

5 Summary of emerging 3DP applications

Aiming to source emerging 3D printed applications and trends, our group (Prof Bhesh Bhandari, Dr Sangeeta Prakash and Dr Fernanda Godoi) attended the BIT's 2nd Annual World Congress of 3D Printing held in Qingdao-China from 13-Nov-2015 to 15-Nov-2015. Professor Bhandari was an invited speaker in the conference and he presented on the topic "Application of 3D Printing for Structuring Healthy Foods". A copy of this presentation is attached.

The conference demonstrated that 3DP technology will be an essential production tool for mass customization which has started remarkably contributing to aerospace and biomedical fields. The benefit of using this technology relies on the free-form design of complex and lighter parts which could not be fabricated through conventional manufacturing processes. In the biomedical area, thousands of people are being benefited by receiving 3D printed implants using as raw material titanium and ceramics. The main advantage can be related to the fact that implants are designed according to the specific characteristics of each human body parts.

Food industry has still to incorporate 3DP as a breakthrough technology. 3DP technology is recognized as essential tool to people interact with food in the future. Digital generation will certainly demand for a digital food. For a decade, 3D food shapes have been successfully printed by using liquid (e.g., melted chocolate, dough and meat puree) and powder (e.g., sugar and chocolate, as fine particles) based materials. Supermarkets are already testing to 3D print customized cakes and restaurants are endeavouring to offer printed desserts. Some even claim that there will be a 3D food printer in every home within the next decade. This is definitely a rapid growing market. *Meat industry has potential to play an important role on the consolidation of this technology based on the rich protein content in the muscles combined with the variety of triglycerides and mineral contents. However, in-depth research is required to convert ideas into reality.*

6 3DP technology: Food design application

Originally, additive manufacturing (AM) technologies were applied for building 3D objects by means of layering deposition of non-food materials, such as, metals, ceramics and synthetic polymers in processes involving the use of organic solvents, extreme temperature conditions or crosslinking agents that do not comply with food safety standards. Therefore, one of the critical challenges in the 3D food printing field has been to align food grade materials with printing processes. Three food materials property related critical factors are suggested here for the rational design of 3D food structures:

- Printability: This feature relies on how the properties of the material enable handling and deposition by a 3D printer and hold its structure post-deposition. The printability of liquid-based AM technologies, such as, drop-on-demand techniques is influenced by the material viscosity or rheological properties. In addition to rheological properties, 3D printing based on extrusion techniques can be affected by specific gelation mechanisms (crosslinking) and thermal properties (melting point and glass transition temperature). Properties like particle size distribution, bulk density, wettability and flowability can also exert influence on powder-based 3D printing;

- Applicability: AM technologies can be attractive by their capability of building complexes structures and textures. In addition, AM becomes more interesting when nutritional value is incorporated to the unique designed structures. The applicability of AM technology is also ruled by the materials properties.

- Post-processing: Ideally, the 3D construct of food should resist to post-processing, such as, baking in an oven, being cooked by immersing in boiling water or deep frying. In the pursuit of a cooking-resistant structures, an accurate selection of materials with appropriate physical-chemical, rheological and mechanical properties are essential.

We emphasize that printability, applicability and post-processing feasibility can be achieved by controlling the physical-chemical, rheological, structural and mechanical properties of the materials (**Fig. 4**). Knowledge of the essential constituents of food (carbohydrates, proteins and fat) and how their properties influence AM technology is critical to guarantee quality in the end-use product.

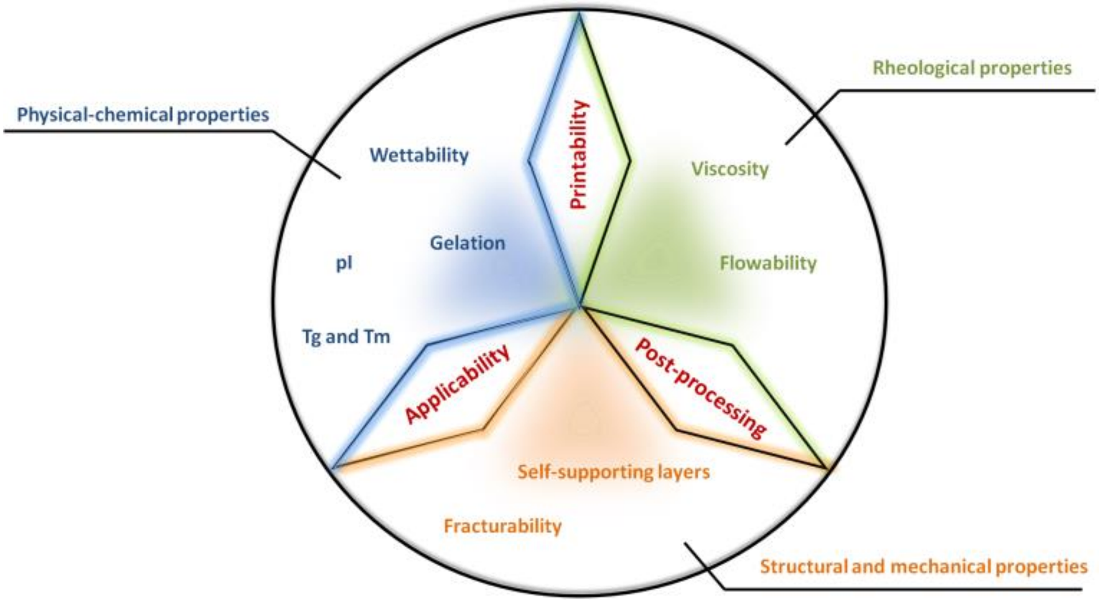


Figure 4: Parallel between materials properties and factors to consider for the rational design of 3D food structures.

6.1 Material supply: liquid and soft-materials

When 3DP technology is undertaken using liquid or soft-materials as ink, there are two methods with potential application on food design: (1) extrusion and (2) inkjet (**Figs. 5a** and **5b**, respectively). In extrusion processes (**Fig. 5a**), formulated ink composed of food ingredients is manually loaded in a cylinder (extruder). The edible ink is extruded out of the nozzle by the force produced by an acting hydraulic piston. The consecutive deposition of layers is undertaken by directing the cylinder at points pre-determined by a 3D model. Depending on the ingredients used in extrusion processes, the binding mechanisms may happen by the accommodation of layers controlled by the rheological properties of the materials, solidification upon cooling or hydrogel-forming extrusion.

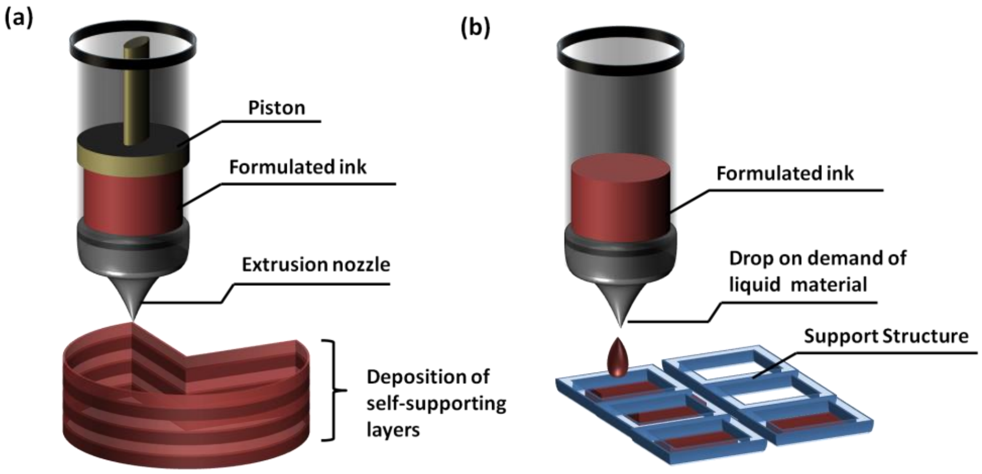


Figure 5: 3DP technologies applicable for food design using liquid or soft-materials as ink: (a) Extrusion and (b) Inkjet.

6.1.1 Extrusion processes

In 3DP technology, extrusion has been applied to print 3D constructs by mixing and depositing self-supporting layers of edible materials such as dough, meat paste, processed cheese and chocolate. Depending on the binding mechanisms involved during the formation of self-supporting layers, extrusion processes can be divided into three groups: (1) robocasting, (2) melting extrusion and (3) hydrogel-forming extrusion; as described below.

(1) Robocasting: Self-supporting layers are created based on the viscosity of the material which needs to be both low enough to allow extrusion through a fine nozzle and high enough to support the structure post-deposition. Rheological modifiers, or additives, can be used to achieve the desired rheological properties but must comply with food safety standards (Godoi et al., 2015).

(2) Melting extrusion: Melted material is loaded into an extruder; 3D shape is formed by the solidification upon cooling of the deposited layers. This method has so far been applied to print chocolate 3D objects, denoting a working temperature which ranges from about 28 ºC to 40 ºC (Hao et al., 2010b, Hao et al., 2010a, Schaal, 2007). The formulation of chocolate self-supporting layers is challenging due to the complex crystallization behavior exhibited by cocoa butter, the main structuring material in chocolate and confections. Six different crystal polymorphs have been identified for cocoa butter (Marangoni, 2003). The correct polymorph should be produced in the chocolate for its best melting, textural and shelf-life properties.

(3) Hydrogel forming extrusion: The extrusion of hydrogel-forming materials is critically dependent on the polymer rheological properties and the gel forming mechanism. At Prior loading into a extruder, the edible polymer solution should present viscoelastic characteristic, and then turn into self-supporting gels prior the consecutive layers are deposited. To prevent premature gelation of the polymer solution inside the printer, temporal control of the gelation mechanisms must be carried out.

Robocasting

Periard et al. (2007) applied robocasting to print cake frosting and processed cheese using the Fab@home Fabrication system (Periard et al., 2007). Using the same system, Lipton et al. (2010) tested a variety of recipes to print sugar cookies. Variations on the concentration of ingredients such as butter, yolk and sugar played an important role to form natively printable dough and resistant on cooking. The authors have also used transglutaminase and bacon fat as additives to make printable scallop and turkey meat-puree, respectively. The resulted meat-based products kept their shape after cooking (Lipton et al., 2010).

Robocasting processes have also been employed by the Netherlands Organisation for Applied Scientific Research (TNO) scientists to print a large variety of foods using essential carbohydrates, proteins, meat purees and other nutrients extracted from alternative sources, such as, algae and insects (Van der Linden, 2015). Most recently, TNO and Barilla (Italian pasta company) have presented the preparation of 3D printed pasta using classical pasta recipes (ingredients: durum wheat semolina and water, without additives) (Sol et al., 2015, Van der Linden, 2015, Van Bommel, 2014). Another example, a company called Natural Machines created Foodini Food printer which extrudes fresh food ingredients to design meals. The extruded ingredients are used for surface filling (e.g., pizza or cookie dough and edible burger from meat paste) and graphical decoration (Chang et al., 2014, Kuo et al., 2014). **Fig. 6** illustrates some examples of 3D extrusion-based techniques applied to print pasta recipe, pork puree and pizza dough.

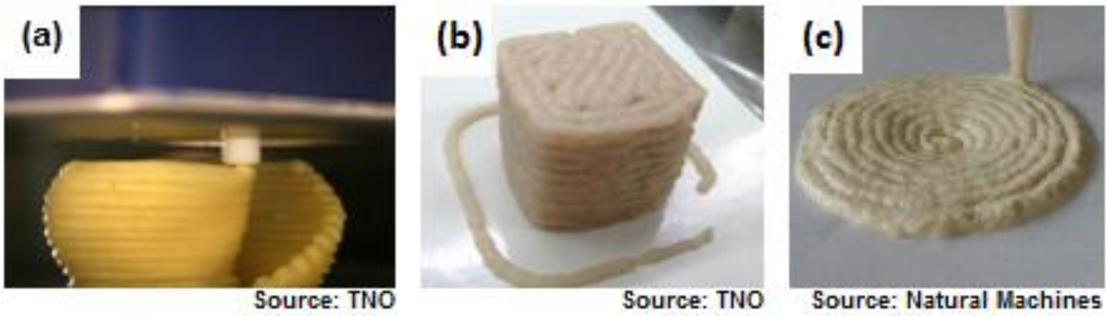
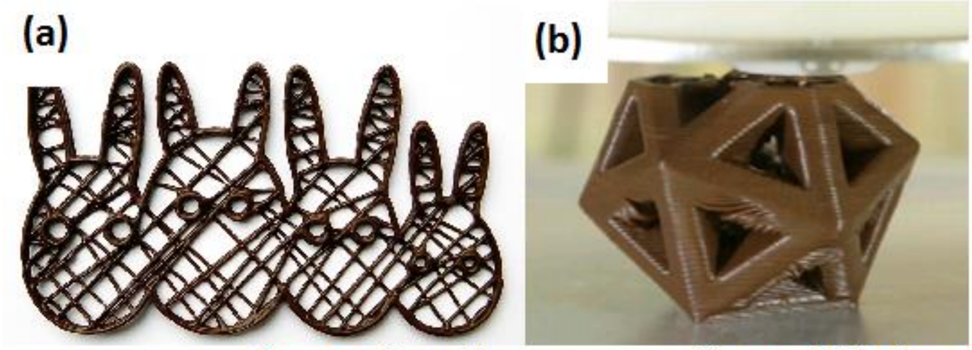


Figure 6: Examples of 3DP technique based on soft-material extrusion: (a) pasta recipe (Van der Linden, 2015), (b) pork puree (Van Bommel, 2014) and (c) pizza dough. Image (c) was reproduced with permission of Natural Machines from data available at https://www.naturalmachines.com/press-kit/#.

Melting extrusion

The chocolate deposition directly into a 3D object by means of melting extrusion was introduced by researchers from Cornell University using a Fab@home Fabrication system (Schaal, 2007). Their studies, however, did not look at the materials properties and geometrical accuracy of the extrudate. Hao et al (2010a,b) revealed the factors influencing the geometrical precision of the chocolate deposition: (1) nozzle aperture diameter, (2) optimum nozzle height from the forming bed and (3) the extrusion- axis movement (Hao et al., 2010b, Hao et al., 2010a). The expertise of the research group led by Hao enabled the foundation of ChocEdge Ltd, a spin-off company from the University of Exeter, which pioneered the commercialization of 3D chocolate printers. **Fig. 7a** shows an example of 3D printed chocolate by ChocEdge. Over the years, many companies applied chocolate extrusion to build 3D objects, for example, Foodini, TNO and recently 3D Systems in partnership with The Hershey Company has introduced the CocoJetTM at the 3D Chocolate Candy printing exhibit (2014) as a breakthrough 3D chocolate printer, enable to build self-supporting layers in a 3D shape, as illustrated by **Fig. 7b** (3DSystems, 2015).



Source: ChocEdge Source: 3DSystems

Figure 7: Examples of 3D printing technique based on melting extrusion: chocolate constructs printed by (a) ChocEdge and (b) 3DSystems (3DSystems, 2015). Image (b) was reproduced from data available at http://www.chocedge.com/creations.php.

Hydrogel-forming extrusion

Generally, the hydrogel-forming mechanisms can be classified in three categories: (1) chemical cross-linking, (2) ionotropic cross-linking and (3) complex coacervate formation (Kirchmajer et al., 2015). Chemical cross-linking is unlikely to be applied for food design, as many cross-linking reagents are harmful and must be completely removed from the designed structure before they are consumed. Conversely, ionotropic cross-linking has been widely applied by food industry, especially in microencapsulation processes (Ching et al., 2015, Bokkhim et al., 2014). As an example, alginate is a polysaccharide composed of mannuronic and glucaronic acid residues (negatively charged at pH values higher than 2) which are cross-linked by calcium ions, resulting in ionotropic gel. A complex coacervate hydrogel is produced when a polyanion and a polycation are bound with one another.

The use of hydrocolloids in combination with food ingredients was reported by Cohen et al. (2009) as an alternative to create printable food materials composed of starch, protein etc. in a platform of different texture and flavours. Testing solely two hydrocolloids, xanthan and gelatin, they simulated a broad range of mouthfeels. The resultant complex coacervate formed by the mixture between xantham and gelatine has shown granularity, which was not observed when the pristine hydrocolloids were tested (Cohen et al., 2009). This behavior can be explained by the hydrogel-forming mechanism of the combination between a polycation (xanthan) and an amphoteric polymer (gelatine). Cohen et al’s study suggests that further materials developments are required to progress in the field of food design using 3D technology. For example, the combination of alginates of different guluronic/mannuronic acid ratios and pectin of high and low degree of esterification has potential to reveal a new printable material for food structure design. By mixing alginate and pectin at low pH values a synergistic gel is form in absence of Ca^{2+} and at high water activity; at this condition, neither of the pristine samples would gel. To promote gel formation, both alginate and pectin chains should be partially positively charged before interacting and methylation is recommended to avoid electrostatic repulsion (Walkenström et al., 2003).

6.1.2 InkJet printing

The technology developed by Grood et al. (2011) for dispensing a liquid onto layers can be classified as InkJet printing (Grood and Grood, 2011, Grood et al., 2013). This technology was commercialized by the name of FoodJet printing and uses an array of pneumatic membrane nozzle-jets which layers tiny drops onto a moving object. The drops together shapes a digital image in the format of a graphical decoration, surface fill or cavity deposition (FoodJet, 2015). Inkjet printers generally handle low viscosity materials; therefore, it does not find application on the construction of complex food structure. Typical deposited materials are: chocolate, liquid dough, sugar icing, meat paste (low viscosity), cheese, jams, gels etc (**Fig. 8**).

Figure 8: Examples of 3DP technique based on inkjet technology: (a) graphical decoration, (b) surface filling and (c) cavity deposition. Images (a), (b) and (c) were reproduced from data available at http://foodjet.com.

6.2 Material supply: powder

After extrusion processes, powder binding deposition is the second most popular system in 3D food printing. This category can be divided into three sub-types: (1) Selective Laser Sintering (SLS), (2) Selective hot air sintering and melting (SHASAM) and (3) Liquid binding (LB); which have in common the powder deposition in bed. By SLS and SHASAM the layers of powder are fused together upon application of a heat source, infrared laser and hot air, respectively. In liquid binding, there is no phase change during layer solidification: a liquid binder is overprinted onto layers of powder that are accumulated consecutively, as in directed fusion (Wegrzyn et al., 2012). Liquid-binding method has been patented as 3D printing (3DP). All three techniques require an additional step for removing the unfused material at the end of construction.

6.2.1 Selective Laser Sintering (SLS)

Although SLS has been extensively used to sinter metals, this technology was also adapted for some food designs. Based on SLS technology, using an infrared laser that heats and sinter the material, TNO has incorporated nutritional value and flavours to powder-based 3D objects. The principle relies on the powder binding as a result of the melting fat and/or sugar of the composition (Diaz et al., 2014b, Diaz et al., 2014a). **Fig. 9** illustrates examples of 3D food objects created by TNO using SLS technique.

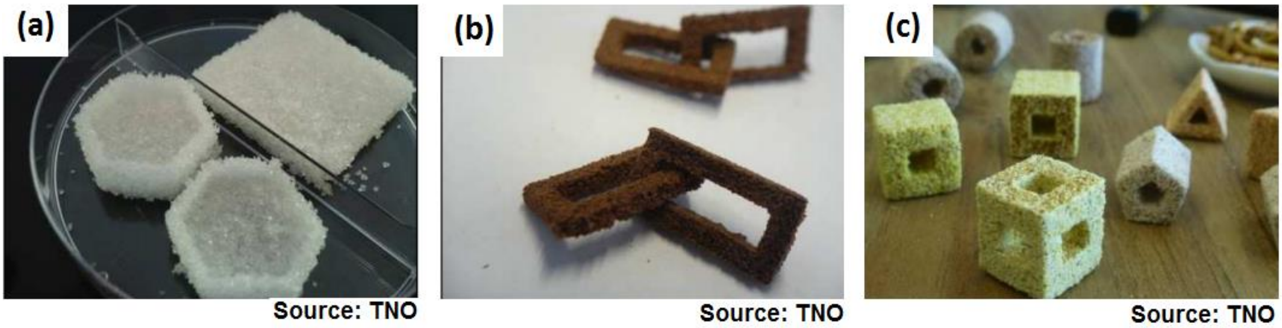


Figure 9: Examples of 3DP technique based on SLS technology: (a) Sugar, (b) Nesquik and (c) Curry Cube, Paprika Pyramid, Cinnamon Cylinder and Pepernoot Pentagon constructs printed by TNO (Van Bommel, 2014).

6.2.2 Binder Jetting (BJ)

An example of BJ technology applied for food design is the 3DSystem's ChefJet printer which uses the Z-Corp inkjet process to produce a broad range of confectionary recipes including sugar, fondant and sweet and sour candy in a variety of flavours-sculptural, as can be seen by the example illustrated in **Fig. 10** (Von Hasseln et al., 2014). Recently, TNO researchers described a liquid binding-based method called Powder Bed Printing (PBP). In this method, edible 3D objects are produced by spatial jetting of food fluid (binder) onto a powder bed containing formulated food powder composed of a water soluble protein and/or a hydrocolloid (Diaz et al., 2015b).

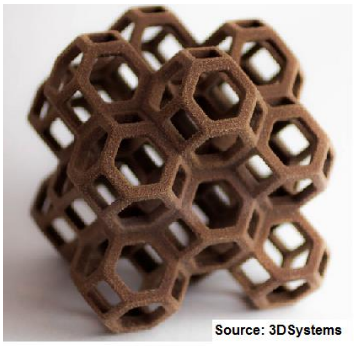


Figure 10: Example of 3DP technique based on LB technology: 3D chocolate structure. The image was reproduced from the data available at http://www.3dsystems.com/culinary/gallery.

6.2.3 Selective hot air sintering and melting (SHASAM)

CandyFab machines, a project by Evil Mad Scientist Laboratories (California, USA), use SHASAM technology to print sugar-based 3D objects. SHASAM technology uses a narrow, directed, low-velocity beam of hot air to selectively fuse together sugar powder, building a two-dimensional (2D) picture out of fused powder. At first, the powder bed is slightly lowered then a thin flat layer of particles is spread to the top of the bed, and selectively fuse the media in the new layer. The freshly printed 2D object is indeed fused to any overlapping connected areas in the previous layer. By performing this step again, a 3D object is gradually built up. Upon finishing the 3D object, the bed is brought to its initial position, disinterring the manufactured model, while the unused powder is kept for use in the construction of the next object (CandyFab, 2006). **Fig. 11** shows an example of 3D structure made of sugar, using SHASAM technology.


Source: Toroidal coil sculpture of sugar by Windell H. Oskay.

Figure 11: Example of 3D printing technique based on SHASAM technology. Image reproduced with permission from Windell H. Oskay, www.evilmadscientist.com.

6.3 Advantages/limitations of 3DP techniques and desirable end-use properties of 3D food structures

Although AM technologies have received a lot of attention in the field of food engineering, the advantages and limitations of 3D printing techniques and their impact on the end-use properties of the materials need to be addressed to exchange, in a profitable manner, the traditional fabrication methods by processes involving AM technology. Ideally, the end-properties related to the mechanical stability of 3D printed food should match with those in conventional manufacturing processes. And, in terms of texture design and nutritional optimization, AM technology would potentially defeat traditional fabrication methods.

Both liquid deposition and powder binding bed techniques are capable to build geometrically complex structures. Liquid-based supplies, however, afford a broad range of materials and mechanisms of binding. Printable mixtures comprised of carbohydrates, proteins and fat can be prepared by tuning the material’s properties, such as, melting and glass transition temperature, gelation and viscosity. The strong interaction between layers is the main factor affecting the stability and self-supporting properties of the final construct. For example, the melting extrusion of chocolate must avoid formation of fissures when the deposited layers are cooled down. Presence of empty spaces between layers (as result of poor interaction) not only cause fracture of the final build but facilitate the undesirable migration of fat (fat bloom) due to the creation of preferential channels.

Powder binding bed techniques have been used to print sugar structures. Especially when heat is used as power source to bind patterns of powder (SLS and SHASAM), the lack of nutritional value in the final product makes this technique less attractive than liquid-based deposition. For this reason, toughness and uniform surface are relevant end-properties of the construct. The powder patterns formed should, in theory, resist fracture upon presence of a crack. The choice of food grade binder can restrict the application of 3DP technique. In addition, weak interactions between powder and binder may lead to rough surface and unstable buildings.

Self-supporting layers, palatability and visual appeal are common desirable end-properties of the 3D food constructs designed via liquid-based deposition, powder binding deposition and bio-printing. Particularly, for the resulting material of bio-printing processes where 3D constructs of meat is designed by depositing biological material (cell cultures), visual appeal would rather relevant than liquid and powder-based AM techniques. This is because bio-printing is an unusual technique in the field of food engineering and the consideration of meat as a post-mortem tissue might cause negative effect on the acceptance of the product for consumption.

6.4 Packaging as a solution for post-processing of 3D printed food

The post-processing of 3D printed edible structures is challenging. When the interaction between layers is weak, the built structure is likely to collapse during cooking into conventional ovens or microwaves. According to Tsakadze and Bardenstein (2013), the use of active packaging can be a solution to preserve the shape of a 3D printed food. They have studied the effect of packaging elements with variable microwave transmission capabilities on the thermal behavior of delicate gel-type rapidly manufactured frozen 3D print food during unfrozen and cooking into a microwave oven (Tsakadze and Bardenstein, 2013).

The packaging units developed by Tsakadze and Bardenstein (2013) for the personalised food were composed of standard aluminium. They had cylindrical shape (Ø90 mm and 20 mm in height) with perforated top; solid bottom and sides **Fig. 12**. Different geometries of perforation were tested; allowing partial penetration of the microwaves through them and gradual heating of the personalised food inside the packaging. The personalised food sample tested consisted of 3-6% jellifying agent Gelea (Biozoon GmbH, Germany) mixed with 100 g of puree made of vegetables, pasta, meats, etc (Tsakadze and Bardenstein, 2013). **Fig. 13** depicts the temperature distribution throughout the cross-section of the food sample and on the top-side using colour codes. Clearly, the geometry presented by **Fig. 13d** provided an enhanced temperature distribution; indicating that by tuning the geometries of the pattern, the amount of microwave energy transferred to the food sample can be controlled. *Current technologies, such as the fabrication of microwave packaging coated with susceptors films, can be used to promote gradual and homogeneous heating of the accumulated layers of ingredients.*

Figure 12: Packaging unit developed by Tsakadze and Bardenstein (2013).

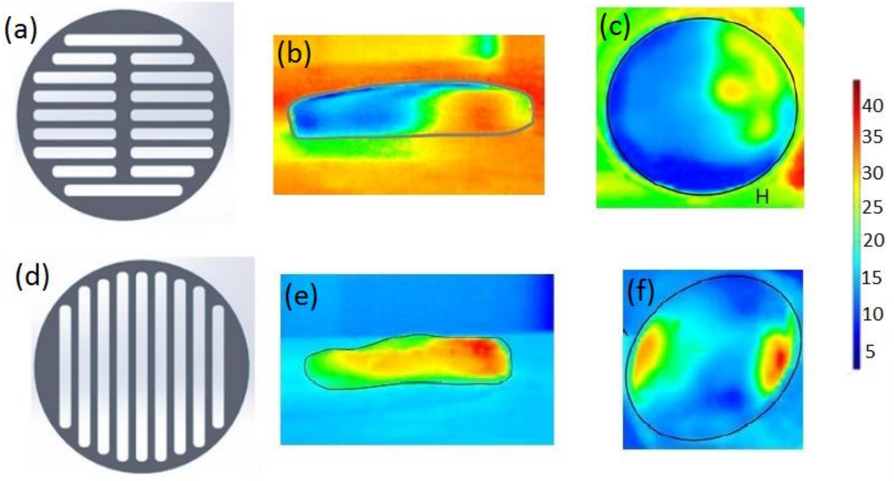


Figure 13: Packaging top perforations (a, d) and their respective temperature profile on the cross section area (b, f) and surface (c, f). Adapted from (Tsakadze and Bardenstein, 2013).

7 Meat science fundamentals to enable 3DP

To enable the application of 3DP technology on meat systems, the research team has reviewed the properties of the main constituents of meat (proteins and fat) and the additives used to enhance texture, flavours and binding of meat pieces in conventional restructuring processes.

7.1.1 Meat proteins and fat

Sarcoplasmic, myofibrillar and stromal proteins

Muscle proteins are generally classified into three separate groups: (1) sarcoplasmic, (2) myofibrillar and (3) stromal proteins; according to their relative solubility at different ionic strengths. Sarcoplasmic proteins are soluble in water or dilute salt solutions (at low ionic strength). The myofibrillar proteins are soluble in concentrated salt solutions, in particular 0.6M KCl. Myofibrillar proteins play a vital role in the muscle contraction which, in turn, provides movement. The stromal proteins, or connective tissues, are insoluble even in high ionic strength solution of sodium or potassium. Based on this feature, they are shown as fibres in the fluidic medium of the body (Wang, 2006). The functional properties of the sarcoplasmic, myofibrillar and stromal proteins have long been recognized in the manufacturing processes of meat products and reformed meat. *By analogy with the potential application 3DP technology, understanding the temperature-induced coagulation of meat proteins is of paramount importance on the construction of self-supporting layers based on meat ingredients.*

Sarcoplasmic proteins are known by their poor binding ability in conventional meat restructuring methods. Despite that, their behavior on heating may affect the characteristics of the surrounding meat elements by coagulating onto them (King and Macfarlane, 1987). *In 3DP, the thermal-coagulating behavior of the sarcoplasmic proteins onto the surrounding components of the build material is likely to facilitate the formation of self-supporting layers. Apart from promoting adhesive forces between the* *ingredients, the gel-like aggregates of sarcoplasmic proteins can enhance the rheological properties of the mixture during accommodation of layers.*

Contrary to sarcoplasmic proteins, myofibrillar proteins are recognized by their ability to bind meat pieces into a cohesive mass when the product is cooked. The potential of myosin, the most abundant myofibrillar protein, in achieving high binding strength leads to toughness during cooking of contracted muscles (King and Macfarlane, 1987). This behavior can be associated to the different gelation mechanisms showed by these two proteins (Tornberg, 2005). The gelation mechanism of myosin involves two steps: (1) aggregation of the globular head portion of the molecule and (2) change in the structure configuration caused by the thermal unfolding of the helical tail of the myosin where hydrophobic groups interact with each other (Samejima et al., 1981, Sharp and Offer, 1992). *Myofibrillar proteins will play an important role in the post-processing of 3D printed meat-based materials. Their ability of achieving high binding strength may hinder the structural and mechanical properties of the meat-construct after cooking.*

Stromal proteins, or connective tissues, consist of collagen, elastin and other insoluble proteins in smaller amounts. Collagen is the principal structural constituent among the connective tissues which includes tendon, bone, cartilage, skin, vascular tissues, and basement membranes. For each type of tissue, collagen assumes particular molecular composition and concentration (Wang, 2006). Collagen has been classified into 28 types of molecules numbered with Roman numerals in vertebrates (I–XXVIII). In addition to the wide range of collagen types, collagen family shows several molecular is forms for a single collagen type and hybrid isoforms composed by the combination between α chains of two kinds of collagen (Ricard-Blum, 2011, Gordon and Hahn, 2010).

Beyond the contribution of myofibrillar proteins for toughness in meat, collagen proteins primarily provide meat toughness due to their lattice-like form of sheets surrounding different levels of the muscle structure. Collagen with fewer cross-links give rise to tender meat. The amount of cross-links and heat stability of collagen increases with animal aging. Heat-labile collagens, from young animals, have their contribution to toughness reduced when the temperature is raised about 50 °C, and is quite small at around 70 °C. Conversely, when the collagen source derives from an old animal, the connective tissue toughness is stable up to 60 °C and remains relatively high at 70 °C.(King and Macfarlane, 1987). *Ideally, the toughness from heat-stable connective tissues must be reduced prior processing meat, regardless the choice of the method to be performed: 3DP technology or conventional restructuring processes.*

Fat

Animal fats are mainly composed of triacyl-glycerols (TAGs) units which are formed by the esterification reaction between fatty acids and trihydric alcohol glycerol. The fat in the muscle and adipose tissue of red meat consists of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). The ratio between saturated and unsaturated fatty acids in the muscle and adipose tissue of meat relies on the animal sensitivity to diet and the time of accumulation (Wood et al., 2004). Differences in fatty acid composition lead to variation on the melting point of meat fat. The melting point of meat fat usually falls within a range of 25 and 50 ºC, as a result of differences in the fatty acid composition. For example, C18:0 melts at 69 ºC and C18:2 n-6 at -5 ºC (Wood et al., 2004).

The levels of saturation together with the length of the fatty acid chain are key factors impacting the melting pointing of animal fat. The trend followed by the melting point is illustrated by the arrows embedded in Table 1. Short-chain fatty acids and high content of double bonds contribute to decrease the melting point of animal fat. Lamb fat shows the highest melting point, followed by beef (tallow) and pork (lard) fat. Another interesting remark is related to the higher levels of unsaturated fatty acids shown by the fat from the subcutaneous adipose tissue. The fat in the outer layers of the animal body tends to be less saturated with a lower melting point. This is because the temperature is low surrounding the body skin and there is a need to maintain the fat in a more liquid state (Brown, 2014).

Table 2 compares typical fatty acid compositions of animal fats (tallow, lard and anhydrous milk fat-AMF) and vegetable oil sources. As noted, animal fats contain high levels of saturated fatty acids which are solid at room temperature. Generally, vegetal oils are high in unsaturated fatty acids and are liquid in nature. As an exception, coconut and palm kernel, showing high levels of C12:0 and C16:0, respectively, are solid at room temperature; however, they tend to crystallize into a brittle non-plastic consistency. Conversely, the solid TAGs in lard and tallow provide plasticity, which is ideal for functionality in bakery systems (Kincs, 1985). Fat droplets are considered surface-active particles and are responsible for stabilising foam in food-grade ingredient systems. In aerated mixes, air bubbles can be stabilized by clusters of fat or, alternatively, foam structure can be formed by aggregation of proteins wrapping fat droplets or bridging protein-polysaccharide after acidification (Truong et al., 2014). *Animal fats and vegetable oils have a potential to be used as additives in 3DP. For the extrusion-based processes where the binding mechanisms rely on the solidification upon cooling; they can be blended or fractionated to tailor the thermal properties aiming to facilitate the formation of self-supporting layers. Fractionated triglyceride (animal or vegetal source) can also act as binder in powder-based 3DP.*

Table 2: Typical fatty acid composition of animal fats and vegetable oils.

Fatty acid	Melting point (ºC)	Animal fats (%)					Vegetable oils (%)		
		Tallow [1] (lamb)	Tallow[2] (beef)	Lard[2] (pork)	Dairy[3]		Coconut[2]	Palm[2]	Soybean[2]
					AMF*	Stearin:Olein			
C10:0 Capric	32	-	-	-	2.90	2.33:3.35	-	-	-
C12:0 Lauric	12	-	0.1	0.1	3.94	3.83:4.34	46.5	0.1	0.1
C14:0 Myristic	53	4.8	2.8	1.4	13.58	14.32:13.40	19.2	1.0	0.1
C14:1 Myristoleic	54.4	-	-	-	1.23	0.87:1.38	-	-	-
C16:0 Palmitic	63	23.4	23.3	23.6	39.40	42.65:36.45	9.8	42.8	10.2
C16:1 Palmitoleic	0	3.5	-	-	1.96	1.37:2.14	-	-	-
C18:0 Stearic	70	15.4	19.4	14.2	11.25	14.39:10.26	3.0	4.5	3.7
C18:1 Oleic	16	40.8	42.4	44.2	23.47	18.32:26.42	6.9	40.5	22.8
C17:0 Margaric	61.3	2.4	-	-	-	-	-	-	-
C18:2 Linoleic	-9	4	2.9	10.7	2.27	1.90:2.23	2.2	10.1	53.7
C18:3 Linolenic	12.8	1.4	0.9	0.4	-	-	0.0	0.2	8.6

[1] BAS, P. & MORAND-FEHR, P. 2000. Effect of nutritional factors on fatty acid composition of lamb fat deposits. Livestock Production Science, 64, 61-79

[2] KINCS, F. 1985. Meat fat formulation. Journal of the American Oil Chemists' Society, 62, 815-818.

[3] TRUONG, T., BANSAL, N. & BHANDARI, B. 2014. Effect of Emulsion Droplet Size on Foaming Properties of Milk Fat Emulsions. Food and Bioprocess Technology, 7, 3416-3428.

*Anhydrous milk fat

7.1.2 The use of additives in meat systems

The use of additives is a common practice in meat systems adopted to enhance the cohesion between meat pieces, water-holding capacity, sensory features (such as, flavour, juiciness, tenderness). In this review, they were grouped into four categories: (1) salts and phosphates, (2) enzymes, (3) hydrocolloids and (4) dairy additives (**Table 3**). By using 3DP technology, the incorporation of high amounts of salts and phosphates can be avoided. The binding of meat ingredients can be achieved by enzymatic crosslinking, gelation or solidification upon cooling as detailed in our article review (Godoi et al., 2015).

Table 3: Additives used in meat systems.

	Functional properties	**Binding mechanisms**	**3DP feasibility**
Salts and phosphates			
NaCl, KCl and phophates	Enhance texture, color and flavour	Electrostatic interactions	Avoided
Enzymes			
Papain, bromelain and ficin (plant source)	Tenderizing	None	Yes
Transgultaminase (animal or microbial source)	Enhance texture, binding, and yield parameters of meat products	Cross-linking	Yes
Hydrocolloids			
Polysaccharides: e.g, alginates, pectins	Thickening, emulsifying and binding agents	pH-induced gelation	Yes
Collagen derivatives	Thickening, emulsifying and binding agents	Heat-coagulation	Yes
Blood plasma proteins	Thickening, emulsifying and binding agents	Heat-coagulation	Yes
Dairy additives			
Whey	Water-holding ability; binding agents	Heat-coagulation	Yes
Sodium Caseinate	Fat and water-holding ability; tenderizing and emulsifying agents	None	Yes

Lipton et al. (2010) have tested transglutaminase as a food additive to build complex geometries out of meat. By incorporating transglutaminase to the meat puree right before printing (extrusion-based 3DP), the material retained its rheological properties, however, a new protein matrix has been developed. Tenderizing enzymes (e.g., papain, bromelain or ficin) can also find applicability on 3DP processes; aiming to enhance the texture of the end-product.

In liquid-based 3DP techniques, hydrocolloids may combine with meat ingredients via gelation mechanism giving rise to a different rheological behavior of the mixture. This approach has been demonstrated by Cohen et al. (2010) as a potential alternative to create a wide range of simulated textures of food using only the combination of xanthan gum and gelatin with flavoring agents (Cohen et al., 2009). In powder-based 3DP techniques, Diaz et al. (2015) have recently reported that hydrocolloids act as a binder component of an edible powder formulation and enhance the control of the migration and flow of the liquid spray into the powder bed during printing. They suggested that the content of hydrocolloid in the powder composition should ideally lie in the range 0.1-2.0wt.%, based on the total dry weight of the composition (Diaz et al., 2015a; Diaz et al., 2015b).

Gelatin protein, a derived protein from the irreversible breakdown of the fibrous structure of the collagen, is a potential candidate for 3DP (Diaz et al., 2015a; Diaz et al., 2015b). Because gelatin dissolves instantaneously in warm water (approximately 40 °C); its use as additive can benefit the preparation of the mixture to be printed. Upon cooling, junction zones are formed by small segments of polypeptide chains reverting to the collagen triple-helix-like structure, leading to gelation (Burey et al., 2008; Ward and Courts, 1977). In addition, gelatin exhibits Newtonian flow in dilute solution except when extended by charged groups. However, the used of flexible protein molecules, such as, gelatin in 3D-extrusion processes must bear in mind that charges on the molecules give rise to relevant effects on viscosity. When both positive and negative charges are present, the molecule is fully contracted at the isoelectric point and a minimum in the viscosity is observed. A change in pH in either direction alters the ionization of the functional groups and increases the preponderance of either positive or negative charges. The mutual repulsion of similar charges extends the molecule and enhances the viscosity. At pH values where the molecule depicts its maximum extension, it may be less flexible and lead to non-Newtonian behavior. The shear rate is another important factor affecting the flow of proteins like gelatin in extrusion processes. Irreversible reduction in viscosity can be observed upon shear application. And, extreme conditions of very high shear rate may lead to a non-Newtonian behavior.

Blood plasma proteins (BPPs), from meat source, are also hydrocolloids with potential use on 3DP processes due to their emulsifier and heat coagulating properties. They can enhance the binding mechanisms between proteins-proteins or proteins-polysaccharides giving rise to stable self-supporting layers.

Dairy additives can also be suitable for 3DP processes. As an example, whey proteins can be used to facilitate the binding mechanisms of meat components by forming gel structures upon heating and pH control. Sodium Caseinates, on the other hand, are not conventionally used to bind meat pieces, but can provide tenderness to the 3D construct built with meat ingredients.

8 3DP in an integrated platform with existing technologies of meat processing

3DP technology can definitely be integrated in a platform with existing technologies of meat processing. The following items briefly describe the fundamentals of the suggested existing technologies which, in combination with 3DP, have potential to add further value to the supply chain of meat.

8.1 Rendering processes

Edibility is defined according to consumer acceptance, demographic distribution, regulatory affairs, economical aspects, hygiene and religious convictions. The production and processing of meat generates inedible raw materials consisting of tissues as hides, skin, hair, feet, heads, bone, blood, organs, glands, intestines, muscle and fat tissues, and whole diseased carcasses. Methods of burial, landfill and composting, incineration or rendering are usually applied to handle and dispose the inedible raw-material from meat industry. Rendering, composting and biogas production have in common the ability to recycle and add to the meat supply chain (Pearl, 2004). Edible rendering plants separate fatty animal tissues into edible fats and proteins. The edible rendering plants are normally operated in conjunction with meat packing plants. Inedible rendering plants are operated by independent renderers or are part of integrated rendering operations.

Edible lard (fat from the clean tissues of healthy pigs) and tallow (fat from the fatty tissues of cattle or sheep) are the most important materials produced from continuous edible rendering of animal fatty tissue. Either the low temperature option or the high temperature option edible rendering processes may be used to render edible fat. The low temperature option uses temperatures below 49 °C and the high temperature option uses temperatures between 82 and 100 °C to melt animal fatty tissue and to separate the fat from the protein. A better separation of fat from protein can be achieved with the high temperature option; however, the protein obtained from the low temperature option is of acceptable quality, whereas the protein obtained from the high temperature option cannot be sold as an edible product (Jayathilakan et al., 2012).

Inedible rendering are classified into two categories: the wet process and the dry process. Wet rendering separates fat from raw material by boiling in water. The process consists of adding water to the raw material and using live steam to cook the raw material and separate the fat. Dry rendering is a batch or continuous process in which the material being rendered is cooked in its own moisture and grease with dry heat in open steam jacketed drums until the moisture has evaporated. Following dehydration, as much fat as possible is removed by draining, and the residue is passed through a screw press to remove some of the remaining fat and moisture. Then the residue is granulated or ground into a meal. The wet rendering process is no longer used because of the high cost of energy and its adverse effect on the fat quality (Jayathilakan et al., 2012).

The combination of existing technologies, such as rendering, with the emerging 3DP technology is a promising alternative to add value to the meat supply chain. This is because rendered lard and tallow are considered powerful additives to optimize the thermal properties of the build material used in extrusion-based 3DP, as discussed earlier in ***Section 7.1.1****.*

8.1.1 Tallow enhancement process

Project A.COP.0067 addressed about two existing technologies and processes to convert beef tallow to a food ingredient with significantly reduced saturation: (1) fractionation and (2) transesterification (Wijesundera et al., 2011).

(1) Fractionation: This technology is used to separate beef tallow into saturated and unsaturated fractions. Fractionation can be achieved either by solvent or dry fractionation processes. Solvent fractionation can yield a major liquid fraction, around

60% by weight of the starting tallow which is completely liquid at refrigeration temperature. This fraction contains only 24% total saturated fat, and a large part of the saturated fatty acids in the liquid fraction is composed of stearic acid which is considered to have a neutral effect on cardiovascular health. *No by-products or trans fats are formed during the fractionation process* (Wijesundera et al., 2011).

(2) Transesterification: in this process tallow is chemically or enzymatically reacted with one or more unsaturated vegetable oils. The saturated fatty acids in the tallow triglycerides are exchanged with unsaturated fatty acids of the vegetable oils to obtain products of reduced saturation. The process allows flexibility of manipulating the end product to desired specifications by the choice of vegetable oil(s) used. This process however, results in the formation of by-products (free fatty acids, mono- and di-glycerides) which need to be removed before the product can be used. No trans fats are generated in this process (Wijesundera et al., 2011).

Unsaturated fatty acids derived from fractionation and transesterification processes can be efficiently incorporated into meat-based formulations. This is a promising alternative to afford meat flavour by using controlled amount of tallow in a healthier trans-free conformation.

8.2 Blood collection and processing

Project P.PSH.0415 (2013) carried out by MLA and Industry sponsor has investigated the production of concentrated liquid plasma (cLP) which is potentially cheaper than currently available spray dried plasma (SDP). To stabilise blood for separation, multiple anti-coagulants were analysed. The pilot scale plasma concentration trials were conducted using membrane technology. **Fig. 14** illustrates the flowchart of the developed process of cLP separation. Notably, the cLP showed a high protein content (MLA, 2013).

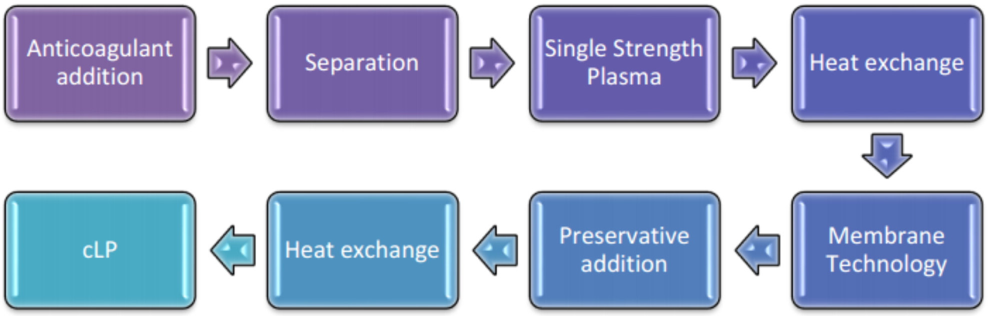


Figure 14: Process flow diagram for production of cLP (MLA, 2013).

Particular attention was devoted to the functionality of blood plasma proteins (BPPs) in the documents A.BIT.0016 and A.BIT.00200. The first document consists of a literature review highlighting the functional properties of BPPs (solubility, emulsifying, foaming and gelation properties) in comparison to a number of protein sources (e.g., from egg and dairy) (Terefe et al., 2011). In the project A.BIT.00200, bovine plasma protein powders produced at pilot scale were selected to evaluate the possible improvement of the bread making properties of low quality plant proteins. They have demonstrated that BPPs can also be used to improve the protein content of low protein plant powders, such as wheat flour and gluten-free flour replacement blends, whilst improving the bread making properties of these low protein plant powders (Glagovskaia et al., 2013).

8.3 Collagen extraction and processing

Alternative uses of hides has been reviewed by Dahm (2011) in the project A.BIT.0011. His study highlighted the existing procedures applied to extract collagen from animal by-products, including skin, bones, and connective tissue. The applicability of collagen-derived proteins, such as gelatin and enzyme-hydrolysed collagen was also explored (Dahm, 2011).

A collagen molecule consists of three left-handed helical polypeptide chains, rolled into a right-handed triple helix with molecular weight of about 300 kDa. Native collagen helices can be unleashed by at least partial denaturation and/or hydrolysis of the triple helix, using chemical and/or enzymatic methods. The latter usually require milder conditions (close to physiological ones) and give rise to products with well-defined properties free of residual organic solvents or toxic chemicals (Kezwoń et al.). Only enzymes belonging to the matrix metallo protease (MMP) family, including various collagenases, which contain both a catalytic site and a hemopoxin domain have the ability of promoting enzymatic cleavage of the collagen triple-helical structure. Heat treatment is often used before enzymatic hydrolysis of proteins to unfold the peptide chain and expose internal cleavage sites (Selvakumar et al., 2012). The major problem associated with enzyme treated collagen is the recovery of collagen free from the enzyme, because the enzyme used for the treatment co-precipitates with the product during the usual recovery procedure, such as salt precipitation (Kezwoń et al.).

Although collagen is generally considered as having relatively low biological value, mainly because of the low amount of branched chain amino acids and lysine (**Table 4**); Hays et al. (2009) have demonstrated that enzyme-hydrolysed collagen protein is equivalent to whey protein as a supplement. They tested the consumption of food supplemented with equal amounts of protein from collagen and whey sources. The nitrogen balance was not different between the two groups, indicating equivalence of both proteins as supplements (Hays et al., 2009).

Table 4: Amino acid composition of the collagen peptides (Zdzieblik et al., 2015).

Amino Acid	Weight (%)
Hydroxyproline	11.3
Aspartic acid	5.8
Serine	3.2
Glutamic acid	10.1
Glycine	**22.1**
Histidine	1.2
Arginine	**7.8**
Threonine	1.8
Alanine	8.5
Proline	12.3
Tyrosine	0.9
Hydroxylisine	1.7
Valine	2.4
Methionine	0.9
Lysine	**3.8**
Isoleucine	1.3
Leucine	**2.7**
Phenylalanine	2.1

A recent study has proved that collagen peptide supplementation in combination with resistance training improves body composition and increases muscle strength in elderly sarcopenic men. The benefits afforded by collagen peptides were related to their rapidly absorption in the small intestine. In addition, collagen contains relatively high amounts of arginine and glycine, both known to be essential substrates for the synthesis of creatine in the human body (Zdzieblik et al., 2015). *Collagen peptides are potential candidates to be ingredients for 3DP extrusion processes. By controlling the level of enzymatic hydrolysis, the viscosity can be tailored. 3DP technologies does not uses extreme conditions of operation, therefore can prevent collagen peptides denaturation: a healthy product can be created.*

8.4 Powdered meat and/or carcass parts

Powdered meat technology was firstly reported by Maclachlan et al. (1924) as a simple method of mixing ground meat with water, pumping-disintegrating operation and, subsequent hot air drying followed by pulverization. The powdered meat normally comprises of an homogeneous mixture of soluble and insoluble substances of the meat formed throughout the pulverization drying process (Maclachlan and Maclachlan, 1924).

Madsen (1983) produced two powdered proteins using offal as protein source from: (1) bacon factories and (2) cattle abattoirs. After heat treatment, the offal is mechanically divided into solid and liquid phases. Protein powder is obtained from both phases. From the solid phase, protein powder is directly generated upon drying of the solids with subsequent removal of the bone pieces. To obtain protein powder from the fluid part, a fat phase is prepared with the liquid which is processed into a size suitable for spray-drying. The powder produced from the liquid phase showed higher protein content and gluing properties which suggest its application as gluing agent or protein supplement to animal food (Madsen, 1983).

Arsem (1991) reported a process for producing freeze dried meat. In this process, ground meat product is slurry-mixed with starch and egg proteins to avoid the formation of oxidation sites on the meat protein. The prepared mixture is heated and kept for a short time at an elevated temperature. Afterwards, the system is freeze dried resulting in slightly soft dried cake with retained meat flavor. The dried cake of freeze dried meat is transferred to a grinder to become powder (Arsem, 1991).

From 2010 to 2015, MLA has reported potential technologies about the production of powdered meat for the development of new snack food products. The document A.MPT.0036 reports the transformation of meat trims into powder using a Gorgens TurboRotor System (**Figure 15**). Three trials were conducted varying the ratio between 90CL trim and hydrolysed bone: (1) trial 1, 90CL trim mill dried at 89 ºC, (2) trial 2, 90CL trim mill dried at 72 ºC and (3) trial 3, 1:1 90CL trim: hydrolysed bone (Dahm, 2014).

The TurboRotor System (**Figure 15**) is composed of a heatexchanger (1) for cooling or heating the process air which is fed into the input area of the Micro-Vortex-Mill (2). In this region, the material is dispersed and homogenised. Consecutively, it reaches the grinding area for simultaneous micronization and drying. The geometry of the grinding tools and stepless variation of rpm determine the size of the particles. The efficiency of evaporation can also be controlled by varying the residence time and turbulence inside the equipment. Before leaving the grinding area, the particulate passes through a classifier area for separation of overs. To sum up, the Micro-Vortex-Mill exerts triple interconnected functions: micronising-mixing-drying. Micronised product is separated from process air by a high-duty cyclone (HFA) (3), high-duty fan with high total pressure (4) and high duty bag filter with low pressure air-jet (5). Clean air leaves the system after passing through a high-duty fan with automatic volume flow control (6). The micronised product is taken at (7) by a collecting screw and directed to a rotational screw (8). It is ready for bagging-off or pneumatic conveying at (9).

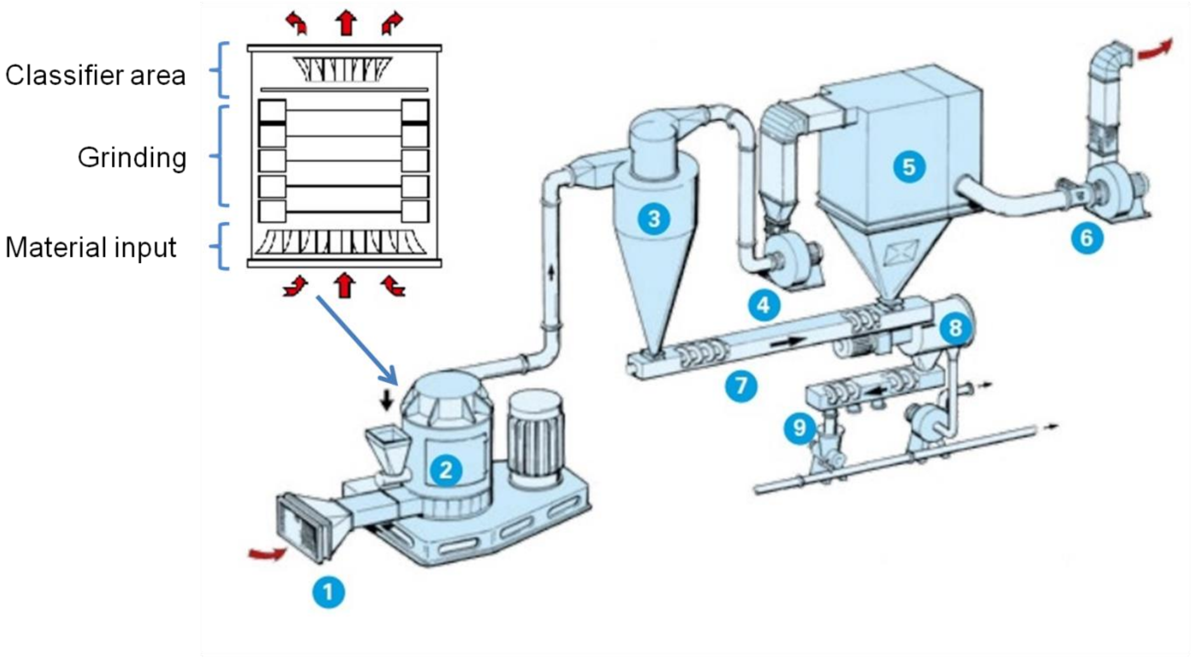


Figure 15: Schematic illustration of the TurboRotor System (Gorgens, 2015).

According to Dahm (2014), the flash-drying provided by the TurboRotor System enable an effective moisture removal (from 0.55 to 0.68 kg water removed /kg feed) and preservation of protein and fat composition (**Table 5**).

Table 5: Meat powder composition using a Gorgens Turbo Rotor mill (A.MPT.0036) (Dahm, 2014).

90CL trim:Hydrolysed bone	Temperature (ºC)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)
1:0	89	3	3.3	2	4.1
1:0	72	2.9	3.1	2.4	3.8
1:1	-	2.1	1.7	1.0	4.2

The meat powder developed by MLA has potential to be used as material supply for 3DP processes. The presence of fat and proteins on its composition can facilitate the binding mechanisms involved during the formation of self-supporting layers. We propose to use the meat powder in the preparation of a slurry-mixture containing additives from animal or plant sources (e.g., fat, hydrocolloids or enzymes).

In a very recent project (A.MPT.0060) a method for producing powdered desiccated liver was described by Sabarez et al. (2015). The main processing operations included cooking in hot water, hot oil treatment and freeze drying, as follows:

- Cooking in hot water: Frozen beef liver cube samples were thawed at 5°C cool room for about 24 hours and then cooked in hot water at 80°C for 15-25 minutes. The cooking process was conducted in a 300 L heating vessel (Cleveland, Canada) with constant stirring to ensure a uniform heating;
- Hot oil treatment: This step was carried out aiming to reduce the extremally high content of vitamin A (fat-soluble) to a recommended safe level for intake as routine consumption of large amounts over a period of time. The cooked/minced beef liver samples were mixed with canola oil pre-heated to the desired temperature of 60°C into a 300 L heating vessel; keeping a proportion of the product and canola oil during the hot oil treatment at 1:2 (w/v). This process was performed continuously for 2 hours with constant stirring. The hot oil treatment was repeated. Afterwards, the oil was drained in a metal mesh to separate the oil from the solid product. Hot water and subsequently centrifuging for 1-2 minutes was applied to remove the remain oil adhered to the product. The solids were spread in thin layers on metal trays and then stored frozen at -18°C for 2-3 days prior to freeze drying;
- Freeze drying: the frozen beef liver samples were freeze dried using a pilot scale freeze dryer (Cuddon FD80, Cuddon Pty, New Zealand) with the capacity of 100 kg of product per batch. The samples were freeze dried for 4-5 days under the standard freeze drying conditions (Setpoint temperatures at -14°C for primary stage 1 and 26°C for secondary stage 2; Vacuum pressure of 2.8 mbar). The freeze dried samples were then milled in Mauri bowl blender for 2 minutes and sieved in 850 microns metal mesh using a SWECO Vibro-Separator (Locker Industries Pty Ltd, Australia) pilot sieving machine. The bulk powder was then blended using a MANCA ribbon blender (Food Industry Products Pty Ltd, Australia, packed and sealed in an aluminium foil packaging, and then stored at 5°C cool room until further final filling/sealing into 15-20 g sachets.

By using the same equipments and conditions described above; beef meat powdering was carried out. Prior drying process, frozen minced beef meat samples were thawed at 5°C cool room for 24 hours and then cooked in hot water at 80°C for 15-25 minutes. The cooked meat samples were then washed with hot water and subsequently centrifuge to remove excess fats adhering to the solids (Sabarez, 2015).

Table 6 describes a comparative nutritional content profile of powdered samples of beef liver, beef meat and these blended samples at the ratio 80:20 (beef liver:beef meat). As expected the vitamin A content is extremely higher in beef liver, in comparison to beef meat powder. Notably, the iron content was substantially higher in the beef liver powder samples; 3 times more than the amount observed for beef meat powder. The content of zinc was relatively high for both samples (Sabarez, 2015). *Powdered beef liver and beef meat are potential candidates to be used as ingredients of printable materials. They can be prepared in a slurry consistency suitable to extrusion-based 3DP technologies. The nutritional content of these powders can be preserved; aiming to deliver a product rich in proteins, iron and zinc.*

Table 6: Nutritional profiles of the final dried powders by Sabarez et al. (2015).

Nutritional content	Powdered products		
	Beef liver	Beef meat	Beef meat: Beef liver (80:20)
Proximates (%)			
Moisture	1.5	1.4	2.6
Fat	21.5	22.4	19.2
Protein	62.3	65.9	71.2
Ash	3.5	1.6	7.1
Carbohydrate	11	9	<1.0
Vitamins (μg/100g sample)			
Vitamin A	**4250**	22	175
Trace elements (mg/kg)			
Iron	160	45	61.5
Zinc	150	180	155
Antimony	<0.01	<0.01	<0.01
Arsenic	0.02	0.032	0.028
Cadmium	0.087	<0.01	0.018
Copper	61.0	2.6	15.0
Lead	0.07	0.013	0.026
Mercury	<0.01	<0.01	<0.01
Selenium	0.48	0.2	0.28
Tin	0.27	0.024	0.048

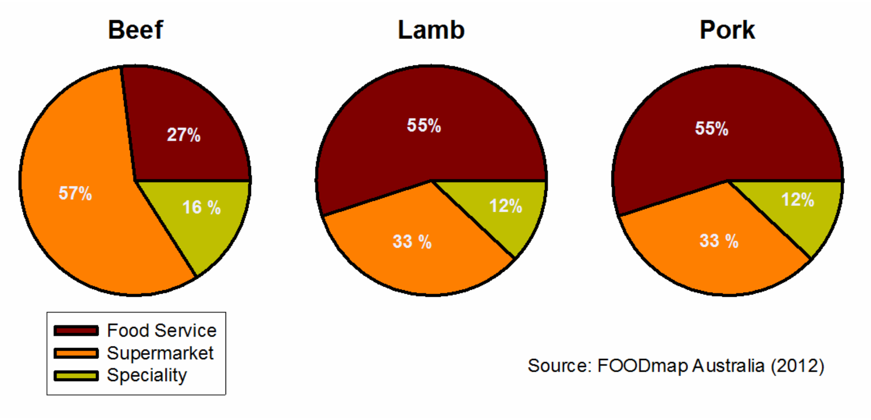
9 Targeting markets for 3D printed food

3DP technology has the ability of producing personalised food. This is the main reason we propose 3DP to attend the needs of food service sector which is defined by the business that make and supply prepared food and beverage products. **Fig. 16** shows the contributions of food service, supermarket and speciality outlets in the domestic red meat market share. A substantial demand for red meat is observed by food service, in comparison with the two other outlets. Based on that, the creation of personalised red meat products in the food sector represents a potential alternative to increase the consumption of red meat and add value for the meat supply chain.

The food service sector is divided in four broad groups: (1) takeaway, (2) dining out, (3) event/leisure and (4) institutional; as illustrated by **Fig. 17**. The outlet numbers are based on information acquired in 2011 by the Australian Department of Agriculture, Fisheries and Forestry. By using 3DP technology to prepare food, essential consumers aspirations pointed by the FOODmap report (2012) can be tackled: sustainability, waste reduction and healthy eating. The topics below explain how:

- Sustainability: 3DP technology enables the creation of tailored nutritional content using sustainable ingredient sources, such as, protein from powdered meat.

- Waste reduction: by creating personalised products, food waste is minimised because the ingredients are mixed according to an individual need. This partly meets sustainability and save money.
- Healthy eating: By 3DP technology, very thin layers of fat, sugar and salt can be deposited during the formation of a 3D shape. This enables the creation of food with enhanced flavours and reduced levels of these ingredients which are constantly associated to healthy concerns such as, heart diseases and diabetes.



Source: FOODmap Australia (2012)

Figure 16: Domestic red meat market share (Department of Agriculture, 2012).

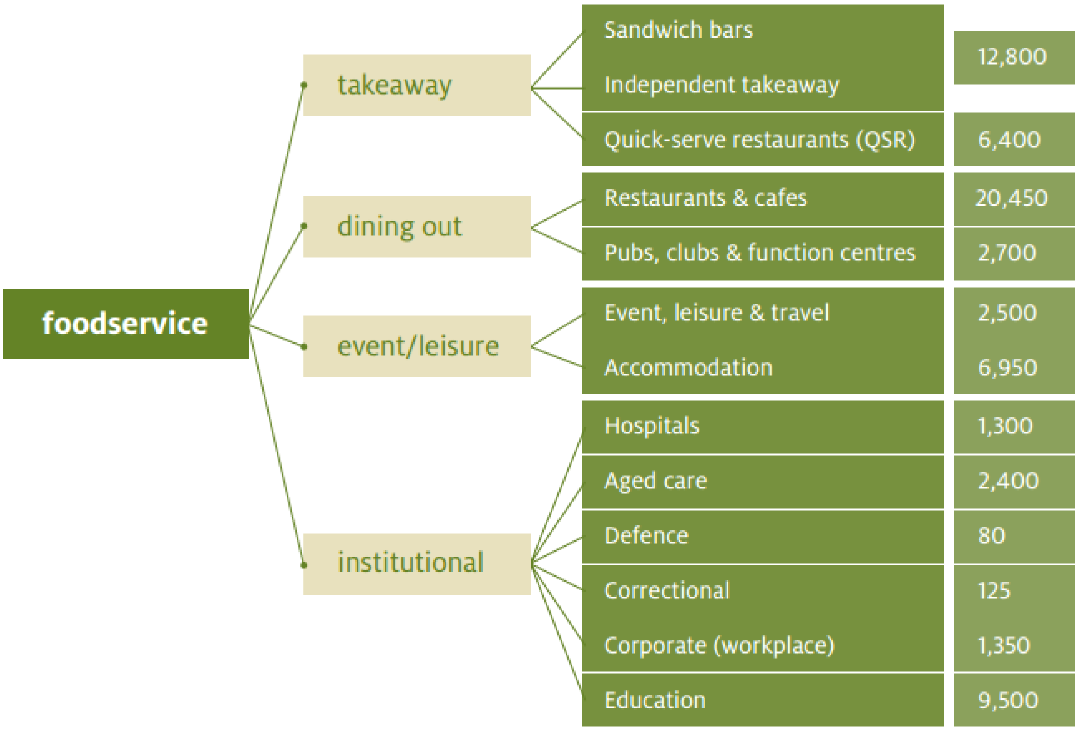


Figure 17: Food service outlet types and numbers (Department of Agriculture, 2012).

Notably, the number of aged care institutions occupies second position of the institutional category. This is due to the accelerated growth rate of the aging population. **Fig. 18** shows that the growth rates of the oldest are estimated to dramatically increase over the next 20 years. The number of people aged 75 years and over is projected to increase by about 4 million between 2012 and 2060 — an increase roughly equivalent to the current population of Sydney. In 2012, there was roughly one person aged 100 years old or more to every 100 babies. By 2060, it is projected that there will be around 25 centenarians for every 100 babies, and with continued small increases in longevity, by 2100, there will be more people aged 100 or more years than babies born in that year. 3DP technology can attend the demand for convenient, healthy and functional restructured food generated by the increase of 'mature aged' consumer segment.

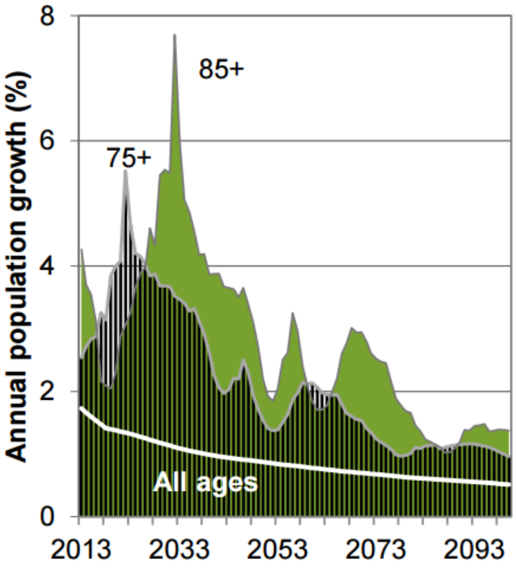


Figure 18: Population ageing until the 22nd century.

10 3DP technology: Packaging application

From our literature review, we have identified two main applications of 3DP technology in packaging industry: (1) tooling confection and (2) active and intelligent packaging.

(1) Tooling confection: Typically, 3DP technology is used for prototyping and short-run manufacturing because of low-volume production and low-duration of tool life in comparison to traditional manufacturing tooling which generally uses metals as raw-material.

(2) Active and intelligent packaging: The use of 3DP technology to produce active and intelligent packaging is considered an emerging technology. In the field of intelligent packaging, we found a report about the design of a smart cap that enables to detect when milk deteriorates. As an example of active packaging, we have also observed that 3DP technology is receiving much attention to fabricate microwavable packaging.

10.1 Tooling confection

Machining molds for packaging industry require substantial time and cost which raise an issue when it is necessary to modify the mold after pilot runs. To overcome this drawback, 3DP technology has been used for prototyping blowing molds, fibre filling molds and thermoforming tools. **Fig. 19** illustrates examples of molds fabricated using 3DP technology. The following items describe the benefits of using 3DP technology in comparison with traditional machining methods (Stratasys, 2015).

- Blowing molds: They are used to fabricate hollow parts (bottles and other containers) by blowing common plastics, such as, polystyrene, polycarbonate (PC) and polyvinylchloride or resins, including polyethylene. Typically, metals are used as raw-material to prototyping blowing molds; the process can be very slow and costly. By using PolyJeT or FDM 3DP technologies the lead time can be reduced by 30 to 70% and the prototype mold cost reduced by 40 to 80%;

- Fibre filling molds: Generally, they are used to mold paper pulp to make containers; trays and other packages Molded pulp packaging tools are normally made by machining a metal tool in the shape of a mirror image of the finished package. Holes are drilled through the tool and then a screen is attached to its surface. The vacuum is drawn through the holes while the screen prevents the pulp from clogging the holes. FDM is an alternative method for producing molded pulp tooling that can provide dramatic time and cost savings and improve the appearance of the finished product. FDM eliminates the need for costly machining of the contour of the tool as well as the holes required to draw the vacuum. FDM also eliminates the need to attach the screen to the mold. FDM molds can be run alongside traditional molds with no alternation to the slurry formula, cycle time, vacuum pressure or other process variables, making it easy to integrate FDM tooling into any molded fiber operation;

- Thermoforming tools: during thermoforming, a plastic sheet is placed above the thermoforming tool and heated until malleable. Then, air is introduced between the tool and plastic sheet improving the consistency of wall thickness across the part. The tool is raised to meet the plastic sheet and the part is formed by pulling a vaccum through the tool to draw the plastic sheet tightly to the surface of the tool. Thermoforming tool can be directly manufactured from the CAD file on an FDM or PolyJet system. Depending on technology and geometry, some finishing work may be necessary for the smoothest possible tool.

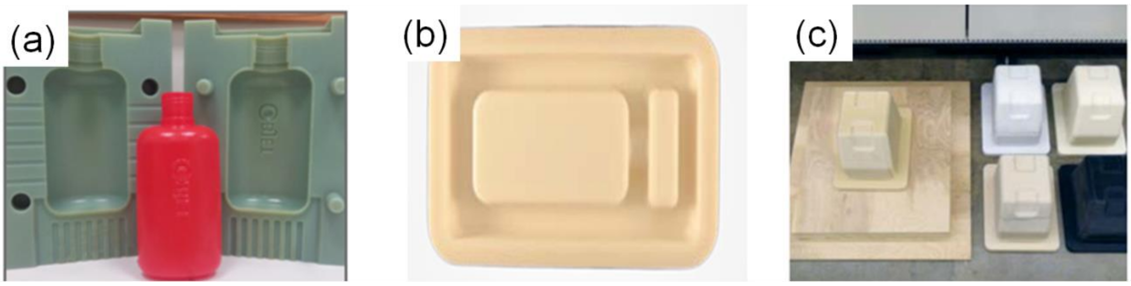


Figure 19: Examples of 3DP application in packaging industry: (a) blowing mold, (b) fibre filling mold and (c) thermoforming tool. Adapted of (Stratasys, 2015).

10.2 Active and intelligent packaging

Certainly, in the future food packages will include radio frequency identification (RFID) tags. RFID tags are responsible for carrying information about products which enable to identify and trace them. They are composed of a microchip connected to a tiny antenna. An external reader emits radio signal to capture data from an RFID tag which is transmitted to a computer. The presence of antennas enlarges the distance that these tags can be read. Based on that, RFID tags shows advantages over conventional bar-codes which need to be in a direct line-of-sight to be recognized by a scanner. Currently RFID tags are used for tracking expensive items and livestock.

3DP technology can facilitate the implementation of complex structures at very low cost. Using an inkjet printer, conductive inks or dielectric materials can be deposited on a substrate, enabling the fabrication of planar circuits, antennas, or sensors. 3D printers make it possible to build polymeric 3D structures with arbitrary heights and thicknesses. Ideally, conductive inks must deliver high conductivity and compatibility with flexible substrates. In general, metal-based inks are composed of metal NPs or metallo-organic decomposition (MOD). NP-based inks, as is self-explanatory based on the name, contain small nano-sized particles of a conductive metal which turns the ink into a colloid suspension. To prevent aggregation and flocculation stabilizing agents can be added (usually a polymeric material). After drying, the conductivity of the ink can be reduced by the formation of insulating layers between the NPs which are created by the stabilizing agent.

The topics bellow describe two examples of 3DP technology applied to fabricate intelligent and active packaging, respectively: (1) “Smart cap” for rapid detection of liquid food quality and (2) Microwave packaging.

10.2.1 “Smart cap” for rapid detection of liquid food quality

Wu et al. (2015) designed 3D-printed microelectronics components and circuit by the combination of 3DP technology and liquid metal paste filling techniques (**Fig. 20**). In this application, as the liquid food (milk) deteriorates, the dielectric constant of the liquid changes and a shift is detected in the resonance frequency of a 3D LC tank embedded in the structure. The dielectric change is monitored wirelessly in real time by observing the resonance frequency shifts of the LC tank via an inductive reader. As such, this smart cap could enable a passive, wireless sensing scheme without the need to open the packages for food safety inspection. Their results indicate that 3D devices with metallic components can open up a new class of applications of 3D printing in the field of smart devices (LC tank: it is an electric circuit consisting of an inductor, represented by the letter L, and a capacitor, represented by the letter C, connected together. The circuit can act as an electrical resonator, an electrical analogue of a tuning fork, storing energy oscillating at the circuit's resonant frequency) (Wu et al., 2015).

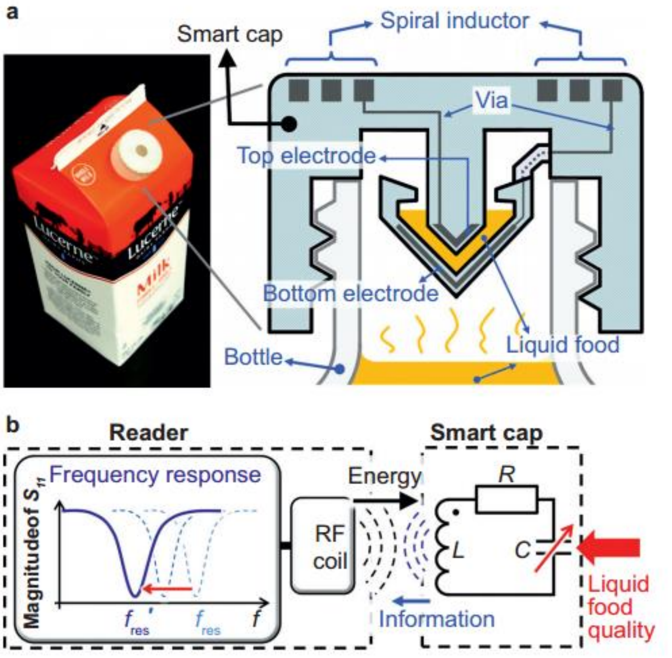


Figure 20: “Smart cap” for rapid detection of liquid food quality proposed by Wu et al.; (a) the smart cap with a half-gallon milk package, and the cross-sectional schematic diagram; (b) sensing principle with the equivalent circuit diagram (Wu et al., 2015).

10.2.2 Microwave packaging

Different from conventional oven, in microwave oven, heat is transferred not only by convection; but by conduction as well. This is because microwave heating is generated by the agitation of water molecules and charged ions exposed to microwaves. The dielectric properties of a food material, along with its thermal and other physical properties, and the characteristics of the microwave electromagnetic field determine the absorption of microwave heating by the food.

The main benefit of using microwave oven is a faster cooking as microwaves directly penetrates into food materials. However, microwaving cannot induce browning or crispness in food because the surrounding air is cold which hampers the water evaporation rate and the development of Maillard reactions. Aiming to make microwaved food resemble food cooked in conventional ovens; susceptors have been used in microwavable food packaging. They are responsible for converting microwave energy into thermal energy to promote preferential heating of food through contact; by this way, crispness is achieved.

Commercially utilized susceptors consist of a metallized active layer (acting as heat source) deposited on a substrate which can be made of paper or plastic. Typically, the growth process of the layer consists of a number of steps: the pre-coalescent stage (which includes atomic surface absorption and critical nuclei creation to a layer thickness of about 5 nm); the coalescent stage (including critical nuclei coalescence,up to about 15 nm); the net creation (chaining of nuclei and their association, up to about 25 nm); the growth of the perforated layer (the layer contains a large number of hollows, up to about 35 nm); and the growth of a homogeneous layer (over 35 nm thick). The process presented can vary slightly in the dependence upon the type of atoms evaporated, the evaporation rate, the residual pressure and the composition in the vacuum chamber, the carrier temperature (usually polyethylene terephthalate, PET) and other growth conditions (Cesnek et al., 2013).

By using inkjet printing technology the susceptor coating can be applied only to the areas pre-determined by a 3D-model. This eliminates the necessity for patterning and removal of deposited materials from undesired areas, decreasing the required materials and waste byproducts. As an example, Babbitt (1991) used electrically conductive graphite particles dispersed into an ink composition to print patterns radiating the desired heat (Babbitt, 1991).

11 Value proposition and next steps

By the end of this study wealth knowledge on the fundamentals of 3DP technology and meat science was acquired. Therefore, the research team has written a value proposition to enable 3DP application on red meat. The research proposal is embedded in the **Appendix III**.

Next steps will involve the optimization of the build material using the following meat by-products in a slurry consistency for application in 3DP extrusion-based processes: powdered meat, collagen derivatives, blood plasma proteins and beef fats (lard and tallow), as mentioned in the **Appendix III**.

Table 7 summarizes the possibilities of using meat as base material in extrusion-3DP. The consistency of meat can be shown as puree (prepared from meat trims) or slurry (prepared from meat powder). The additive choice will determine the binding mechanisms during the formation of self-supporting layers; being classified into three groups: (1) solidification upon cooling, (2) gelation, ionic crosslinking and (3) enzymatic crosslinking. The emulsifying properties of some additives, such as, sodium caseinate, can help the development of the mentioned mechanisms.

Table 7: Additive's choice and their respective afforded binding mechanisms between layers.

Additive	**Binding mechanisms**
Lard/tallow	• Solidification upon cooling
Blood plasma protein	• Gelation, ionic cross-linking • Emulsifying properties
Collagen derivatives	• Solidification upon cooling and/or • Gelation, ionic cross-linking
Transglutaminase	• Enzymatic cross-linking
Hydrocolloids	• Emulsifying properties • Gelation, ionic cross-linking

12 Discussion

Project V.RMH.0034 encompassed all the key aspects involved to undertake successful 3DP of meat products, including functional properties of meat constituents (fat and proteins) and the effects caused by the incorporation of additives. This study has brought insights to the efficient use of meat by-products. Our value proposition emphasizes that 3DP can be conducted in an integrated platform with existing technologies of meat by-products processing. Several previous projects developed by MLA (e.g., A.BIT.0011, A.MPT.0035, A.MPT.0060, P.PSH.0415, A.BIT.0015 and A.BIT.00200) would profit from the adoption of 3DP technology as strategy to add value to meat and livestock industry, as detailed in **Section 8**. The specific project objectives were efficiently achieved. The following topics describe the extent to which specific project objective was met.

Describe science fundamentals on how 3D printing works in both food and packaging applications

We efficiently conducted a literature review about 3DP technology applied to food design. 3DP is considered an emerging technology which gives freedom to design innovative edible product in a variety of shapes, flavours, nutritional content and texture. Our study was submitted to the Journal of Food Engineering. Due to the core capabilities of the research team, 3DP application on packaging was explored in less extent than the application on food. The brief review performed on the packaging field revealed that 3DP technology has also brought innovation to packaging industry. 3DP technology has been used for prototyping blowing molds, fibre filling molds and thermoforming tools. In the field of intelligent packaging, a smart cap, fabricated using 3DP technology, has enabled to detect when milk deteriorates. As an example of active packaging, we have also observed that 3DP technology is receiving much attention to fabricate microwavable packaging using metal-based conductive inks.

Complete brief literature review and patent search to identify emerging areas for this platform

Critical literature review was conduct together with a patent search. Our patent search revealed 81 patents describing methods to print 3D structures. With 64 patents, the liquid-based techniques represent the majority of the 3DP processes (**Appendix I**). Identification of emerging 3D printed applications and trends was also achieved from the participation on BIT's 2nd World Congress of 3D Printing, Qingdao China. This conference has broadened our knowledge on 3DP technology by showing different softwares and applications. After participation on a number of forums discussing about the hot topics in 3DP, new thoughts were developed to conduct research on 3D printing of meat.

Present overview of global applications and network of providers in Australia

While there are no providers of 3D food printer in Australia, **Appendix II** presents a list of global providers and applications of 3DP technology on food sector.

Describe meat science fundamentals to enable 3D printing

Meat science fundamentals were reviewed based on the functional properties of the main constituents of meat (proteins and fat). This knowledge was pursued to help understanding the binding mechanisms involved between layering deposition when meat ingredients are used as supply material. The existing meat by-products technologies was also studied and related with the functional properties of meat constituents. This approach aggregated a strong scientific background which enabled the composition of a value proposition for MLA stakeholders.

Define the Value Proposition for red meat and 3D printing and recommended next steps

The value proposition was composed aiming to achieve efficient utilization of meat by-products. The research proposal involves the use of 3DP in an integrated platform with existing technologies for meat by-products utilization (**Appendix III**).

Meat as build material: What is the ideal 3D printer?

Throughout the project V.RMH.0034, the knowledge acquired from MS1a (the principles of 3DP printing technology), MS1b (emerging 3DP applications and trends) and MS2 (fundamentals of meat science and conventional processing used to transform meat by-products) enable to answer this question:

At the moment, the suitable 3PD technology on red meat can be based on extrusion deposition. This is because meat products are rich in proteins with heat-coagulating and emulsifying properties which can contribute to the formation of self-supporting layers. Lard (low melting point) and tallow (high melting point), by-products of the meat industry, are considered excellent ingredients to tailor the thermal properties of the build material. *The 3D print "ink" can be prepared from meat protein solution in combination with hydrocolloids from different sources (e.g. animal, dairy, plants, alginates) or slurry-mixed with the meat powder developed by MLA (benefits will be delivered for A.MPT.0036 and A.MPT.0060).*

Apart from fact that meat powder has potential to be used as material supply for powder-based 3DP technologies, such as, Selective Laser Sintering (SLS), the high value of a powder-based 3D printer would make the process unfeasible economically. A SLS printer can cost up to $300,000. Besides, there is no 3D printer available in the market to print food powder. SLS for food materials is at research level only, as the examples of 3D printed powder-food developed by 3DSystems and TNO (Godoi et al., 2015). The meat powder (project A.MPT.0036) has potential application as "ink" for liquid-based 3D printers; where it can be reconstituted to slurry, dough or paste consistency and extruded together with different additives, such as, enzymes, fat (tallow or lard) and hydrocolloids (e.g., blood plasma proteins and collagen derivatives).

13 Conclusions/Recommendations

The specific objectives of this project were successfully achieved. We reviewed the fundamentals of 3DP technology, meat science and conventional meat processing technologies: restructured meat, extruded meat, powdered meat and rendering fat. The applicability of meat ingredients in 3DP technology was explored by analogy with their functional properties in conventional meat systems.

Based on the knowledge acquired throughout MS1 and MS2, we state that meat ingredients have great potential to be used in the formulation of 3DP inks. Aiming to achieve efficient utilization of meat by-products a value proposition was composed. This proposition involves the use of 3DP in an integrated platform with existing technologies for meat by-products utilization.

V.RMH.0034 will help MLA to support future investment decisions and undertake ground breaking research about 3DP technology using meat as ingredient. 3DP technology was recognized as a powerful approach to add value to the meat supply chain because of meat by-products can be used to create new textures and flavours.

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