

Trends in Hybrid Cultured Meat Manufacturing Technology to Improve Sensory Characteristics

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Abstract

The projected growth of global meat production over the next decade is attributed to rising income levels and population expansion. One potentially more pragmatic approach to mitigating the adverse externalities associated with meat production involves implementing alterations to the production process, such as transitioning to cultured meat, hybrid cultured meat, and meat alternatives. Cultured meat is derived from animal stem cells and undergoes a growth and division process that closely resembles the natural *in vivo* cellular development. Cultured meat is emerging as a widely embraced substitute for traditional protein sources, with the potential to alleviate the future strain on animal-derived meat production. To date, the primary emphasis of cultured meat research and production has predominantly been around the ecological advantages and ethical considerations pertaining to animal welfare. However, there exists substantial study potential in exploring consumer preferences with respect to the texture, color, cuts, and sustainable methodologies associated with cultured meat. The potential augmentation of cultured meat's acceptance could be facilitated through the advancement of a wider range of cuts to mimic real muscle fibers and products through 3D scaffolding. This review examines the prospective commercial trends of hybrid cultured meat. Subsequently, the present state of research pertaining to the advancement of scaffolding, coloration, and muscle fiber development in hybrid cultured meat, encompassing plant-based alternatives designed to emulate authentic meat, has been deliberated. However, this discussion highlights the obstacles that have arisen in current procedures and proposes future research directions for the development of sustainable cultured meat and meat alternatives, such as plant-based meat production.

Keywords: Cultured meat, Alternative protein, Hybrid cultured meat, Scaffolding, Texture, Color, Muscle fiber.

Introduction

According to the OECD/FAO (2021), it is anticipated that the consumption of meat proteins worldwide will experience a 14% rise over the next decade, mostly driven by escalating income levels and population expansion. This projection is in comparison to the average consumption levels observed between 2018 and 2020. The escalating demand is intricately linked to a range of issues, encompassing public health and environmental challenges, as well as concerns over animal care and ethical considerations. It is anticipated that the rise in global meat production would lead to a significant increase in Greenhouse gas (GHG) emissions (FAO, 2014), as well as water pollution, diminished access to fresh water, and adverse alterations in biodiversity, all of which pose a direct threat stemming from the expanding livestock sector (Steinfeld, et al., 2006). According to Sarlio (2018), the utilization of calories by animals for maintenance and non-edible tissue creation is estimated to be approximately 97%, which is widely regarded as inefficient. The consumption of meat especially red meat in excessive amounts has been found to pose health hazards for individuals (Abu-Ghazaleh et al., 2021). Nevertheless, cultured meat (CM) from live animal cells may fit the consumer demand for growing real meat consumption (Ismail et al., 2020).

CM is produced from live animal cell cultures and is considered an alternative to real meat (Post M. J., 2014). CM approach acquired a larger concentration globally in the research arena, media, investors, specific groups of consumers, and animal welfare organizations (Goodwin & Shoulders, 2013; (Schneider, 2013; Verbeke, 2015; Stephens et al., 2018). A great number of efforts are ongoing to produce CM to mimic real meat from beef, poultry, pork, and seafood by researchers. The so-called hybrid CM (HCM) is a hybridization of animal cells and possible ingredients from plants, bacteria, and algae including various types of binders to mimic the real meat taste and texture of real meat (Lee S. , et al., 2023). Hybrid meat products represent a

novel category of goods wherein a portion of the meat content, often around 20%, is substituted with alternative protein sources, including plant-based alternatives (Baune et al., 2023). According to existing research, these products have the potential to function as a viable option for a particular demographic seeking to reduce their meat consumption, therefore aiding in the transition towards a diet that is both healthier and more environmentally sustainable.

CM is expected to be safer in terms of public health (Willett, et al., 2019) and a way out toward a sustainable way to produce food with high-quality protein and theorized to be able to produce 1 billion beef burgers from a single cow biopsy (Kumar, 2021). The first-ever regulatory approval for cultured meat production was for “Eat Just” in 2020 in Singapore (GFI., 2020). Upside Food and GOOD Meat got final approval from the FDA and USDA for marketing and selling cultured chicken meat products in 2023 (GFI, 2023). CM still possesses major challenges in terms of high production cost, product quality, and consumer embracing (GFI, 2023). Numerous developments need to be addressed to make cultured meat mimic real meat e.g., myoglobin production, marbling of the meat, culture media optimization, sustainable production techniques, muscle bundle formation to mimic different meat cuts, major sensory attributes, nutritional values, and scaffolding technology (Shira et al, 2022), etc.

Several commercial manufacturers ventured into plant-based meat (PBM) market and few stand out as pacesetters e.g., Beyond Meat, Impossible Foods, Eat Just, Memphis Meats, Aleph Farms, Mosa Meat, Cultured Decadence, and BlueNalu (GFI, 2023). These companies advance toward sustainability in manufacturing HCM, including PBM, through production capacity, product quality, legislation, and research and development based on consumer demand. In this regard, this paper reviews the opportunity of HCM or meat alternatives and the advanced

challenges related to the structural and sensory characteristics to focus the consumer sustainability.

The commercial prospect of cultured meat

According to the Good Food Institute, the Overall value of CM in 2022 is estimated at USD 0.3 Billion and is expected to grow to USD 20 Billion in 2023 with a growth rate of 143% (GFI, 2023). Health safety, environmental pollution, and vegan movement issues trigger a driving force to alternative meat solutions both by the industry and the consumers. Table 1 indicates an immense opportunity for this sector in the future (GFI, 2023). There are 28 companies working on meat-based products, out of which sixteen are involved in the production of cultured meat or plant-based analogs and scaffolding. There is one research-based company working on serum-free culture media and the other one on techniques and ingredients to boost the texture of meat alternatives. Nine companies are in the development phase for the production of CM and meat alternatives and one is involved in the development of a cell line for CM.

In recent days the intensified interest of consumers in PBM products and CM is an outcome of continuous product development and marketing efforts. The conception of any new invention always has some drawbacks and, in this case,, there are challenges from consumers and technological barriers (Sanchez-Sabate & Sabaté, 2019) especially on meat color, texture, taste, etc. as real meat and emphasized by researchers for future improvements (Weinrich, 2019; Bakhsh, et al., 2021; Bakhsh, et al., 2022). By embracing and developing various sorts of reorganizational technology, such as 3D scaffolding, PBM and CM may be able to deliver safer and more environmentally friendly state products. So far plant and animal-based cultured meat found to be safe and could be ideal alternatives to address the growing need for animal protein

with the growing global population (Zhang et al., 2022). In the parts that follow, an explanation will be given of the technological characteristics that might offer solutions to the problems in Human Capital Management (HCM). This is done with the intention of generating the need for additional research.

Sensory characteristics of hybrid cultured meat

Processing of different functional ingredients turns into a brand-new appearance with consistent nutritional characteristics known as 'Restructured food' (Polášek, et al., 2021). Various food additives are included to develop compounded structured food (Carpentieri et al., 2022). Structured meat is a type of reconstituted food that is attaining admiration from consumers as a substitute for orthodox meat products. HCM is a kind of structured food that reassembles animal-based CM with PBM to create a new texture and appearance that mimics real meat. Structured meat like HCM is considered to be safe, healthy to consume, and capable of solving animal welfare issues. However, in order to gain a preference that can meet consumer expectations, HCMs containing PBM will have to have sensory characteristics similar to those of natural meat (Bakhsh, et al., 2022). The texture, color, taste, etc. would be the major criteria in marketing this kind of cell-based CM in the future (Joo, et al., 2022). In addition to these sensory characteristics, consumers will also be increasingly interested in nutrition. The development processes employed in controlled atmosphere (CA) packaging are intricately linked to the structural composition of muscle tissue, as the shape of muscles is directly associated with the nutritional and sensory attributes of meat. Satellite cells are predominantly obtained from the skeletal muscles of several animal species, including cattle, chicken, swine, lamb, and fish, in contemporary methodologies. The fibrous texture commonly observed in red meat, such as beef, can be attributed to the intricate hierarchical tissue organization. The muscle fiber serves as the fundamental functional component, encompassed

by connective tissue, intramuscular fat, vascular, and nerve tissues. According to Listrat et al. (2016), the key factors that influence muscle texture and quality characteristics are muscle fibers, fat, and connective tissue. HCM products have the advantage of being able to produce customized nutritional products for consumer needs. In this regard, research on the development of HCM products focused on the nutritional and structural development of meat that meets consumer needs will present immense opportunities for CM market in the near future.

To effectively design HCM products that align with customer demands, it is imperative to do research on plant proteins that constitute a substantial proportion of HCM and/or PBMs produced using these proteins. While the PBM sector is experiencing growth in the market, it is important to consider that negative sensory feedback from customers has the potential to restrict its market expansion. The texture and sensory attributes of plant-based meat (PBM) are not on par with those of actual meat, as the fibrous structure of meat plays a vital role in this regard (Lee S et al., 2023). The inclusion of hydrocolloids is known for their potential to improve the textural properties of PBM. Hydrocolloids could be precious for the development of PBM to improve texture and sensory characteristics. The myofibrillar protein (MP), which primarily comprises myosin and actin, is crucial in the development of the desired texture and water-holding capacity (WHC) of comminuted beef products. High Acyl gellan gum was shown to produce large fibers when PBM containing soy protein isolate (SPI) (Taghian et al., 2023). Positive feedback was revealed from the addition of wheat gluten on the texture, binding properties, moisture retention, and sensory attributes in PBM (Chiang, et al., 2021). But as an allergen wheat gluten is not accepted by consumers with enteric problems (Theethira & Dennis, 2015).

Meanwhile, edible cell microcarriers can be directly added to the final HCM product, which may reduce the cost and yield losses (Nienow et al., 2014). Edible cell carriers like Chitosan-

collagen (90:10) were successfully produced to culture from different primary livestock animal cells and have considerable potential for the development of cultured meat products when incorporated these carriers can contribute to the CM sensorial and nutritional values (Anton et al., 2022).

Plant proteins used in manufacture of HCM and/or PBM

The obstacles encountered in the development of plant-based meat substitutes revolves around the replication of tactile characteristics inherent in meat-based products, such as mouthfeel, chewiness, cohesiveness, and springiness. Nevertheless, the development of desired characteristics in food products relies on the interplay between the selected protein(s) and/or non-protein components, as well as the utilization of sophisticated and innovative techniques, owing to the inherent disparities in the structural chemistry of plant and meat proteins (Mattice and Marangoni, 2020). Various plant proteins, including those found in cereals, legumes, pulses, and leaves, can be used to make plant meat alternatives (Lee S. , et al., 2023). Commonly wheat, corn, rice, barley, sorghum, and amaranth grain are sources of cereal proteins. Moreover soybeans, rapeseed/canola, sunflower seeds, sesame, flaxseeds, and linseeds are sources of oil-seed proteins (Kyriakopoulou et al., 2018).

Soy protein, specifically in the forms of soy protein isolate (SPI), soy protein concentrate (SPC), and soy flour, serves as the primary plant-based protein utilized in the production of PBM and/or HCM. Soy proteins have a wide range of functional qualities, including hydrophilicity, lipophilicity, emulsifying capabilities, and the capacity to create gel structures. In order to produce artificial meat substitutes, soy protein concentrate and soy protein are

frequently employed and they play an important role in reducing product cost and making it competitive in the market (Bakhsh, et al., 2022).

The extraction process demonstrates that SPI possesses a comparative advantage over SPC in terms of protein content. Various approaches were employed for protein extraction in the case of each product. An alcohol extraction is used to collect SPC where it retains about 70% of its protein content (Lee S. , et al., 2023). On the other hand, protein retention is 90% in the case of SPI. An alkaline extraction and leached at an acidic pH assisted in having 90% of its protein content. SPI is well accepted in the industry and research arena for the production of meat analogues due to its brighter color and a drearier flavor than other soy-sourced ingredients (Kyriakopoulou et al., 2018). The end products of SPI possess a meat-like texture after hydration and are much more economical than the other plant protein sources (Sun et al., 2021). The physiological and nutritional characteristics of soy protein have been found to surpass those of actual meat. Meat mimics created employing soy-derived proteins exhibit a larger protein content compared to authentic meat, and furthermore, they possess elevated nutritional properties. Moreover, it has been demonstrated that the fat and cholesterol levels in plant-based alternatives are comparatively lower when compared to those found in traditional meat sources (Cavallini et al., 2006).

Wheat gluten, which forms thin proteins on elongations and may be swiftly converted into a fibrous protein, is another often used grain protein. Wheat gluten used as a scaffolding material in the development of meat analogues in its 3D network form. Wheat protein is known for its, which helps provide the most commonly encountered form of consistent meat analogues. Additionally, to create meat extenders, wheat gluten can be combined with soy flour or soy protein isolate (Asgar et al., 2010). The allergens in wheat gluten should be taken into account while using in the production of meat analogues, as these could be problematic for children

(regular and with special need) and sensitive consumers (Keet, et al., 2009; Martínez-Villaluenga et al., 2008). Nevertheless, Due to its water insolubility, wheat gluten cannot be used extensively in the food processing industry (Asgar et al., 2010).

Legumes are widely recognized for their high-quality and abundant protein content. Legume protein sources encompass a variety of plant-based foods, including beans, chickpeas, lentils, lupines, and peas. Legume-based proteins have a notably abundant amino acid composition in comparison to alternative plant sources. They are rich in lysine and threonine, but in comparison to cereal grain-based proteins methionine, cysteine, and tryptophan are shallow (Kurek et al., 2022). Emulsification, gelation, and foam stability are three functional characteristics exhibited by legume proteins, which enhance the efficacy of using plant protein products as food additives. Emulsification, gel formation, and foam stabilization are three functional traits of legume proteins that improve the utilization of plant protein products as food additives (Ettoumi et al., 2016). High-moisture extrusion has been used to create materials with a fibrous morphology from pea protein isolate and wheat gluten (Schreuders et al., 2019). Soy protein isolate (SPI), soy protein concentrate (SPC), and other plant proteins derived from soybeans are often used ingredients in the production of plant-based meat (PBM). The identification and incorporation of alternative sources of plant-based proteins is of utmost importance. Furthermore, apart from using single ingredients, the combined effect of one or more ingredients is necessary to produce significant combinations suitable for commercial production, economic feasibility, and overall sustainability (Bakhsh, et al., 2022). The primary issues associated with plant-based proteins pertain to their eating quality, specifically their capacity to effectively replicate the appearance, texture, flavor, taste, and nutritional composition of actual meat products. Furthermore, it is imperative that the product possesses the capacity to be manipulated in a manner akin to uncooked meat, and then prepared in a manner resembling meat cooking techniques, resulting in a final product that elicits a sensory

encounter like to the consumption of meat. Despite advancements in the enhancement of texture and flavor in plant-based meat alternatives, there remain obstacles in delivering a satisfactory sensory encounter and enhancing the nutritional value of such products.

Improvement of sensory characteristics

Improving the meat color

In natural meat, the presence of myoglobin is what gives it its distinctive red hue; in cultured meat, on the other hand, myoglobin is not present, which results in a more pale appearance. Myoglobin expression is extinguished at ambient oxygen conditions (Thorrez, et al., 2006; Gholobova, et al., 2018; (Lee et al., 2022)). A number of initiatives guided way out to increase the myoglobin content in CM or PBM (Fraeye et al., 2020), summarized in Table 2.

Improving marbling

Consumer preference starts with the optical inspection of a product and has immense importance in meeting their expectation, especially for novel foods (Post et al., 2020; Moore et al., 2021; Post & Hocquette, 2017). The presence of marbling patterns in meat is known to increase consumer acceptance when purchasing (Lo Surdo et al., 2013). This can be achieved by mimicking the marbling patterns found in traditional meat. By incorporating fat cells alongside muscle cells, cell-based meats can develop marbling patterns that enhance their visual appeal. Additionally, advanced 3D printing technologies can be utilized to create intricate structures that resemble marbling patterns, further improving the visual appearance of cell-based meats (Shujian, et al., 2021). To create a well-integrated structured CM that replicates the texture of conventional meat, the engineered adipose tissue was integrated within engineered bovine muscle tissue using a gentle stitching process that allowed the co-culture of

the integrated construct while preserving the delicate mature adipocytes (Zagury et al., 2022). Marbling and three-dimensional type superior structure in cultured meat yet not established in current culture techniques (Jurie, et al., 2007; Datar & Betti, 2010).

Improving structure by scaffolding

The process of CM creation necessitates the utilization of satellite cells derived from live animal muscle, which are subsequently deposited onto a biomaterial substrate commonly referred to as a scaffold, microcarrier, or film. This foundation stimulates the proliferation of satellite cells in order to increase their population size. Following this, the progression of muscle fibers and adipose tissue formation persists through the process of differentiation, with the ultimate objective being the replication of the authentic structure of meat (Ostrovidov et al., 2014).

In the field of cultured meat, it is of utmost importance to ensure the appropriate arrangement of fiber bundles in order to accurately replicate the structural characteristics observed in conventional meat. The attainment of well-organized meat can be accomplished through the utilization of scaffolds, which provide cells with an extracellular matrix-like framework for the purposes of support, differentiation, and proliferation. Different strategies are in use for scaffolding e.g., microcarriers, porous scaffolds, fiber scaffolds, hydrogels, 3D printing, scaffold-free approaches, etc., and synthetic polymers, self-assembling peptides, ECM molecules, plant & fungus derives materials are commonly used scaffolding materials (Bomkamp, et al., 2022). There are four key aspects that need to be taken into account for tissue engineering (TE), of which the mechanisms are similar to scaffolds being used for CM production. Biocompatibility, biodegradability, the architecture of the scaffold, and the technology used to manufacture the scaffold are the four important areas that need to be addressed during the production of CM through tissue engineering (Seah et al., 2022). Micro

carriers found to significantly increase the efficiency of cell proliferation and differentiation and it's achieved by expanding the surface area and serving anchorage-dependent cells a link (Jong, 2023). The properties of scaffolding such as microcarriers have often been inspired by the field of TE, which needs to be adapted for CM (Singh, et al., 2023).

Scaffolds that are frequently employed has the ability to be utilized in the production of human cardiac muscle (HCM) because to their edible nature or their rapid biodegradation during the phases of differentiation and maturation. The utilization and advancement of edible scaffolds have the potential to decrease the expenses associated with the production process of HCM, as these scaffolds can be commercialized as a product. According to Ng and Kurisawa (2021), if edible scaffolds are to be retained in the final product, it is imperative that they are both harmless and safe for consumption, without altering the flavor or texture. Further investigation is necessary to explore sustainable, efficient, viable, and ultimately consumable polymers for implementation in the cultured meat sector.

Conclusion

The allocation of investments and the level of interest exhibited by investors in this particular industry may serve as a promising indicator for the prospective expansion of cultured meat and meat alternatives in the future. Consumer interest in meat alternatives, such as CM, may experience a decline unless their prices become comparable to that of actual meat. The primary research and industrialization objective should revolve around replicating the color and texture of authentic meat in order to align with consumer preferences and enhance pleasure. The consideration of production costs is a crucial aspect to be taken into account throughout the development of a product such as lab-grown meat, specifically in the context of HCM manufacturing. The development of non-animal-based colorants, additives, and colloids that

are more efficient and cost-effective, along with the implementation of sustainable scaffolding techniques for constructing 3D scaffolds and ingestible scaffolding material, holds the potential to revolutionize the meat industry. This vision is shared by researchers and industrialists who aspire to establish a sustainable future for the industry. The application of the electrospinning technology has the potential to be utilized in the development of cost-effective 3D scaffolds, hence potentially leading to a reduction in the overall production cost of cultured meat. Furthermore, there exist potential opportunities for enhanced alignment of muscle fibers in a three-dimensional scaffold, thereby replicating the authentic structure of meat.

Conflicts of Interest: The authors declare that they have no conflicts of interest.

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Table 1. A list of companies focused on the development and production of cultured meat, plant based meat, technological advancements, and support supplies.

Company	Focus	Present Phase	Year Founded	Animal-Type/Analog	Country
Aleph Farms	Cultivated beef steak	Production	2017	Beef/veal	Israel
BioTech Foods	Cultivated meat products	Production	2017	Beef/veal	Spain
BioBQ	Cultivated beef brisket and jerky	Production	2018	Beef/veal	United States
Gaia Foods	Cultivated meat products	Production	2019	Beef/veal	Singapore
GOURMEY - Suprême SAS	Restaurant-grade meats directly from animal cells	Production	2019	Duck	France
Ivy Farm Technologies	Cultivated pork products.	Production	2019	Pork	United Kingdom
Lab Farm Foods	Cultivated meats, including pork and chicken nuggets	Production	2019	Pork/Chicken	United States
Alife Foods	Cultured schnitzel (special meat cut)	Production	2019	Beef/veal	Germany
Steakholder Foods	3D printed cultivated meat	Production	2019	Beef/veal	Israel
Mirai Foods AG (fmr. AlphaMeats)	Cultivated meat products	Production	2019	Beef/veal	Switzerland
Blue Ridge Bantam	Hybrid alternative poultry products	Production	2020	Chicken	United States
Novel Farms	whole cuts of cultured Iberian pork	Production	2020	Pork	United States
Ohayo Valley	Plant-based and cultivated meat to produce cultivated wagyu ribeye	Production	2020	Beef/veal	United States
Magic Valley	Cultivated lamb meat	Production	2021	Mutton/lamb	Australia
Ambi Real Food	Cultured beef based meat products	Production	2021	Beef/veal	Brazil
LiquiBio	Edible scaffolding	Production	2022	Beef/Chicken/Pork/Turkey/Duck/Mutton/Goat	United Kingdom
Joes Future Food / Nanjing Zhouzi	Serum-free culture media	Research	2019	Pork/Beef/veal	Mainland China

Meat.The End	Techniques and ingredients to boost the texture of meat alternatives	Research	2020	Chicken/Beef/veal	Israel
SuperMeat	Cultivated chicken	Development	2015	Chicken	Israel
Appleton Meats	Clean ground beef, chicken	Development	2016	Beef/veal/Chicken	Canada
Uncommon	Cultivated meat	Development	2018	Beef/veal	United Kingdom
Mission Barns	cultivated meat (Kosher Bacon)	Development	2018	Pork	United States
SciFi Foods	Cultivated and cultured meat products	Development	2019	Beef/veal	United States
WildBio (Formerly Mogale Meats)	Cell-lines for cultured meat	Development	2020	Beef/veal	South Africa
Ants Innovate	Cultivated whole meat cuts	Development	2020	Pork	Singapore
Re:meat	Large-scale production of cultivated meat	Development	2022	Beef/veal	Sweden
MyriaMeat	100% real meat muscle	Development	2022	Beef/veal/Pork	Germany
Foodurama	Plant-based and cell-cultured meat.	Development	2022	Beef/veal/Chicken/Mutton/lamb	Indonesia

Source: (GFI, 2023).

Table 2. Summary of observations from recent research on developing plant based or tissue culture meat color and concerns to trigger future research.

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Present observations	Future research needs
Low oxygen ambience may increase myoglobin expression during culturing muscle fibers (Post & Hocquette, 2017) (Simsa, et al., 2019).	Need further evaluation to assess the impact of low oxygen conditions on cultured meat (Shane B, et al., 2009) (Schlater et al., 2014).
Hypoxic conditions resulted better efficiency with increased glucose uptake and lactic acid production and Lipids or acetic acid media showed stimulation of myoglobin expression (Matilda et al., 2015).	Need further evaluation to assess the impact of low oxygen conditions on cultured meat (Shane B, et al., 2009) (Schlater et al., 2014). Possible acidification may damage the cells (Isam et al., 2015).
Sufficient iron is mandatory in the cell for myoglobin synthesis and color development (Natalie et al., 2019).	Uptake is transferrin-dependent (Isam et al., 2015).
Myoglobin content in cultured meat may be improved by direct inclusion of myoglobin, recently addition of metmyoglobin (the oxidized form of myoglobin) increased cell proliferation and the content of myoglobin in cells (Simsa, et al., 2019).	Bioavailability of iron and inclusion limit into myoglobin needs to be studied (Simsa, et al., 2019). Still, the color of cultured beef meat is pale than real beef after adding metmyoglobin.
The possibilities of artificial color or plant-derived colors are under observation but only for processed meat products. Soy leghemoglobin was able to give the color and taste of a real beef burger (Watson, 2019).	As red meat is associated with health concerns, it is wise to develop alternative colorants for cultured meat (Gamage et al., 2018).

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