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Cultured meat technology: an overview

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1 Introduction

With the introduction of the cultured meat concept, the past decade has seen the remarkable rise of a new scientific field based in the food technology domain, with the potential to change our current food systems[1]. Cultured meat is made from animal cells grown outside an animal. The principal approach behind producing this food is based upon the idea to use cell and tissue culture techniques, originally developed for the medical science field, to grow all components of edible meat *in vitro*. At a minimum, this comprises muscle cells, which grow and develop into enlarged cell assemblies. Ideally, this biomass would feature as many aspects of animal-derived meat as possible.

Briefly, the technology entails obtaining a biopsy from a live animal and growing it to the desired volume in a suitable production environment with the help of specific media to feed the development. For the creation of three-dimensionality and texture, typically a bio-scaffold is incorporated at some point during the process (Fig. 1). The resulting material is meat, and it will have to be labelled as such. Genetically, cultured meat is identical at the cellular level to conventional meat, and in the public debate, it is also

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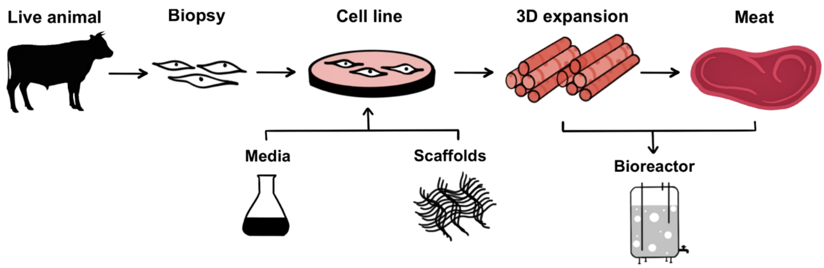

Figure 1 Overview of the cultured meat process. Initial cells are sourced via a biopsy taken from the animal of choice. Subsequently, from this source, relevant cell types are selected to develop an immortalized cell line. Finally, by using enriched media, these cells are prepared to proliferate and differentiate in a bioreactor with or without the addition of scaffolding material, which provides texture to the growing material.

referred to as lab-grown meat, clean meat, or *in vitro* meat. The scientific and commercial domain dedicated to this innovative technology refers to itself as 'Cellular Agriculture' (cellAg) to highlight the idea of introducing new domestication, i.e. the scaled production of cellular material that so far had only been obtained by higher animals or plants[2]. The term cellAg was coined in 2015 by the New Harvest non-profit organization for animal products made without animals. The technology holds the potential to improve global food security by addressing major ethical, environmental, commercial, and public concerns.

A key objective behind any industrial effort in the context of food is to provide food security. The emerging global food security gap necessitates the production of additional 60–70% calories by 2050, assuming a population of about 10 billion people globally[3]. With the intent to alleviate the burden on current food systems and to improve nutritional quality and animal welfare, the field of cellAg offers a potential solution, albeit technologically challenging to accomplish.

The technology will provide significant advantages as compared to conventional animal-based meat if the product categories are chosen well. Based upon the enclosed nature of growing biomass in bioreactors, all ingoing and outgoing material streams can be controlled.

Still, by far the largest challenge remains scaling of the technology at an affordable cost to provide nutritious, safe, and affordable material in large and impactful quantities. Current setups and approaches are nowhere near the output of the current animal-based livestock industry.

A wealth of conceptual papers, commercial reports, and anticipated consumer scenarios are being published, although we still do not know how CellAg will live up to expectations and how the related products will be perceived. The current situation is that products will be available and even

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are available in minute quantities to boutique restaurants and specialty niche outlets.

Cultured meat products are being envisaged at various levels of sophistication. At the simplest level, unstructured cellular material will be available that can be diluted with water to provide for a broth or soup stock. With increasing complexity, more texture can be introduced and minced meat or 'ground meat-like' structures will be available, material which at this stage already should contain additional cells or tissues such as adipocytes to provide for the organoleptic property of fatty taste or texture as well as the nutritional benefits of these cell types. Chicken nuggets or any artificially shaped meat products from other species belong in this group as well and might be available without additional cell types. The most ambitious product form will be analogous to a meat cut. To achieve meaningful meat cut analogues, it will take considerable time and significantly more technological development.

The field of food science is full of examples where one material is supposed to mimic another one, from tofu-based vegetarian meat in the tenth century Song dynasty to margarine, which is designed to mimic butter, and to specific molecules that are developed to mimic the taste of salt or sugar, such as aspartame, which imparts a sweet taste. In the cultured meat domain, it will be interesting to see if mimicking animal-based materials will be the ultimate ambition or if the derived materials will succeed to attain a food category by themselves. Why mimic animal material if you want to abandon animal-based materials?

To make cultured meat a success story, it is not just a matter of significant advances in one technology; success is dependent on making strides in an entire technology suite if the resulting products are not just meant to serve as a gimmick in a niche market.

2 Limitations of conventional food systems

Most of the food eaten by humans is derived through some agricultural and/or industrial process. Often conventional food systems are associated with deforestation, monocultures, and greenhouse gas (GHG) emissions, followed by energy consumption and emissions from transport and retail (Fig. 2). The principles of current agricultural food production are based upon the 'green revolution', which led to a significant boost in productivity post World War II[4]. A global transition took place over the past 60 years towards our current food systems, which rely on monocultures, policy, machinery, and chemical inputs, such as fertilizers, antibiotics, and pesticides. The aim behind these developments has been to satisfy the need for a more affordable, safer, and rapidly growing food supply.

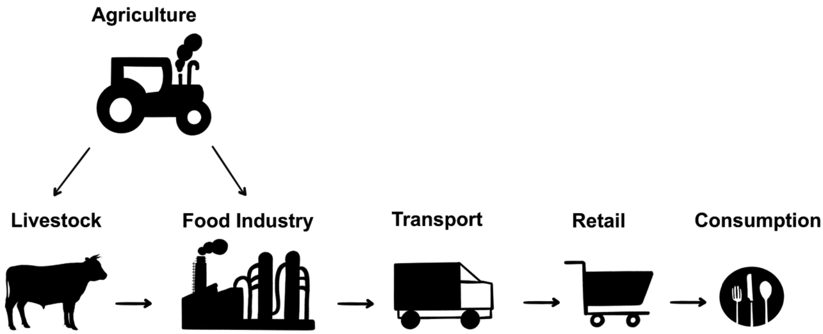

Figure 2 All stages of a food system are vulnerable to being broken, thus threatening food security. Agricultural boundaries include deforestation, GHG emissions, monocultures, resistance to pest and weed, and pollution of air and land. Vulnerability issues are exacerbated in livestock farming and the food industry. The overall sustainability of a classical food system can be improved by the responsible development of cellular agriculture.

However, as the global population increases and other challenges such as climate change occur, there is a need to address the environmental, social, and economic impacts of current agricultural technologies. We have come to a point where agricultural practice can even present a health risk – since the 1940s, nearly half of all zoonotic diseases in humans have been found to have come from livestock. Of similar concern is the usage of antibiotics in the production of meat, which has been linked to a rise in antibiotic-resistant pathogens[5]. The latter incidences are closely related to the industrialization of animal agriculture, which has increased accessibility to animal proteins and cheaper products. It also has created a perfect niche for zoonotic transmissions and has established itself as a threat to global food security.

The effects of industrial agriculture can be categorized into environmental, social, and economic impacts.

2.1 Environmental impact

Current farming practice increasingly is ecologically unsustainable. The intensive farming necessary to produce the high yields of nutrition that support global food needs has led to increased environmental impact. This includes higher use of fertilizers, irrigation, and land expansion. Our industrial agriculture successfully satisfies food demand and brings economic benefit but impacts the environment. Deforestation, pest and weed resistance, soil degradation, destruction of natural habitat, and water pollution are some of the major drivers of climate change. Emissions from the livestock industry can be categorized by source. Enteric fermentation, manure management, feed production, and energy consumption are the four main processes that contribute to GHG emissions (Fig. 3).

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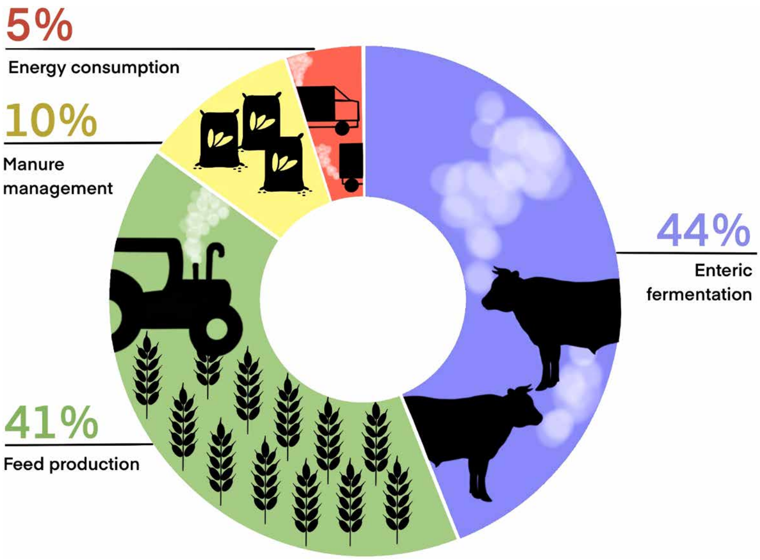

Figure 3 Global greenhouse gas emissions from livestock supply chains based upon 2015 data[30]. Enteric fermentation accounts for 3.5 gigatonnes CO_2-eq for the methane generated from ruminants and non-ruminants. Manure management includes methane and nitrous oxide released during the anaerobic decomposition of manure, which accounts for 0.8 gigatonnes CO_2-eq. Feed production accounts for 3.3 gigatonnes CO_2-eq from feed crop expansion and fertilizer production. It also includes nitrous oxide from nitrogenous fertilizers and use of manure in crop fields. Energy consumption accounts for 0.4 gigatonnes CO_2-eq attributed to the entire supply chain system, including crop management, processing, transport of feed, production site, milking, packaging, and product transport.

Millions of hectares of land have been dedicated to growing monoculture crops to produce livestock feed. This contributes to deforestation, soil degradation, and the increase in the use of herbicides, insecticides, and fertilizers. Hundreds of millions of tonnes of manure are produced every year to sustain livestock. Often, fields that rely on manure, store it in large open lagoons which upon flooding contaminate the soil and water systems with antibiotics, bacteria, pesticides, toxic chemicals, etc. A significant factor regarding climate change is the production of methane by rumen digestion of cattle, particularly cows (Fig. 3). Regarding heat accumulation, methane is a 25-times stronger effector as compared to carbon dioxide[6]. A wide range of issues are linked to the production of meat, and there is great interest in potential solutions. This includes the use of sustainable feed[7] or feed that limits greenhouse gas emissions[8] as well as clear incentives for the industry to reduce environmental impact. However, implementing these changes will take time and requires strict compliance. It is conceivable that cellAg could avoid or help limit a range of these emissions. The degree to which the change to cultured meat will have an impact does depend on how the industry improves key metrics such as power generation from renewable sources to drive bioreactors, or diverse mixtures of plant materials to serve as a supply chain. If unchanged, the environmental impact of current food systems will intensify with the growth of the meat industry as the population increases to the predicted 9.8 billion by 2050.

2.2 Social impact

To ensure global food security, a sustainable food system is required. Animal products such as meat, aquaculture, eggs, and dairy use about 83% of the world's farmland and contribute to about 57% of the different food-based emissions, while providing only 37% of our protein and 18% of our calories[2]. In addition to other environmental matters, there are increasing ethical concerns about a variety of traditional agricultural practices such as the use of 'factory farming'. Factory farming has been indicated to place undue stress on animals and may also have a role in promoting disease[9]. A recent study indicated that consumers have begun to link factory farming with cruelty and the potential for new zoonotic diseases[10]. While the social impact of a transition to cellAg products such as cultured meat is still being studied, there is a clear emerging consumer desire for non-animal-based products reflected by the rise of alternative meat and milk products[11].

2.3 Economic impact

The livestock market contributes 40% to the global agricultural output value and supports approximately 1.3 billion people in terms of food security and livelihood[2]. This reality is supported not only by consumer desire but also by governmental policies. An example of this is the lower cost of animal feed owing to government subsidies with profit margins being significantly lower if these subsidies are removed[12]. Further, tax exemptions are provided over other supplies such as equipment and employment structures. The current framework does not consider these externalized costs and the impacts on public health and on natural resources are not considered as well. These hidden costs of the current agricultural model need to be considered when modelling the transition to cellAg products. The emerging concept of 'true cost' is helpful in illustrating the overall picture[13].

It is not the first time the food industry needs widespread innovation. To meet food demands, already during the industrial revolution small farm layouts were developed into intensely packed breeding grounds for livestock. Another boost to productivity occurred during the green revolution in the second half of the twentieth century[4]. Raising livestock for food has its limitations. For example, in the US, the average number of days for chicken to reach the consumer has been reduced from 112 to 48, while the average weight has increased from 1 kg to about 3 kg[12]. To meet the growing food demands of an increasing human population, continued efficiency improvements are required[12]. The consumption of meat is engrained in all cultures and may have been responsible for shaping human development as well as providing significant nutrition and pleasing taste to consumers[14]. However, with the ever-increasing demand on land, water, GHG emissions, and ethical concerns, alternative solutions should be considered. It is a goal of the nascent cellAg field to best address as many of these points as possible.

3 Rationale for a transition to cellular agriculture

Conceptually cultured meat is an extremely appealing way to address the current shortcomings of the industrial livestock production system. So far, alternative meat products, some of which are based on recombinant and plant proteins, have already started replacing animal-based meat. The same will likely be observed for cultured meat as the field develops and the general public better understands the technology. Some current studies have indicated reluctance in the population to try cultured meat products[15,16]. Most respondents do support further research into the field[15,16] to improve a range of concerns such as worries about the taste or quality of the final cultured meat product. As the field develops, it is hoped the production of meat can be more sustainable without animal suffering and at a realistic economic return[17,18].

Driven by the limitations of animal-based meat, there have been advancements in plant-based meat as well. Other than the conventional plant-based alternatives such as tofu, novel plant-based meats are developed with enhanced meat-like sensory characteristics. Both plant-based and cultured meat consider animal welfare, human health, greenhouse gas emissions, land and water conservation, and economics to provide an effective alternative to traditional agriculture. Both types of alternatives have their benefits, and neither should be positioned to solely monopolize the market. However, cultured meat has the advantage of being sensory and nutritionally equivalent to conventional meat. It will be key to keep an eye on its attributes as pilot plants are getting bigger and production facilities are being implemented.

Importantly, advances in the cellAg field remain the subject of controversial debate, and this debate is conducted with occasionally sobering views on both sides[19]. Some clarity is provided via two studies commissioned by the Good Food Institute to be conducted by CE Delft. In a Techno-Economic Analysis (TEA) for the production of cultured meat, the overall feasibility and chances for economic success are carefully laid out[20]. In a prospective Life Cycle

Assessment (LCA) study, the same group investigates the environmental impact of a cultured meat industry operating at commercial levels[20].

As with all innovation, it will be of the essence to drive technology development in the cellAg space with rigorous and independent scientific exploration. This can be achieved by providing sufficient time and funding to ensure the sustained market introduction of safe and nutritious cultured materials. Moreover, some of the targeted health benefits might convince non-meat consumers or picky eaters to eat cultured meat if a viable nutritional option is provided without the involvement of livestock animals[21,22].

3.1 Health

Cultured meat products are being described as holding great potential to improve health and nutrition. This idea remains to be proven and will likely require further research, particularly as more products are developed. As with all dietary principles, it will be important for consumers to adhere to a balanced diet. There is a possibility to replace unfavourable saturated fatty acids with healthier alternatives, such as omega-3 fatty acids[23]. Other modifications such as inclusion of dietary fibre from cell scaffold materials or direct addition of nutrients such as vitamins could lead to cultured meat being a healthier choice than traditional meat[23]. Cultured meat is intended to be free of antibiotics, which are heavily used in intensive farming. Animal-based food production is an incubator for antimicrobial resistance, and about 70–80% of antibiotics worldwide are used on farm animals[24]. Cultured meat presents the opportunity to produce meat lacking pathogen contamination and environmental contaminants such as antibiotics, microplastics, and heavy metals like mercury in fish products. A Harvard study published 90 reasons to consider switching to cultured agriculture[24]. The study rationalizes the use of cultured meat as it would reduce antibiotic resistance, zoonosis, and GHG emissions and how this could result in nutritious clean meat. Apart from zoonosis, various abattoirs and meat packaging plants across the globe have been a risk factor throughout the COVID-19 pandemic, which occasionally has been attributed to poor hygiene conditions[25]. Without high-density animal farming, zoonotic outbreaks like swine and avian flu might be less prevalent.

3.2 Sustainability

By definition, sustainable food meets the need of the current population without compromising future generations. Cultured meat could meet the growing demands of 10 billion people by 2050, and it can have a positive impact on global hunger and climate[26]. Moreover, it has the potential to meet climate goals without affecting consumption patterns by sustainably emitting less

GHG compared to conventional meat[27]. In terms of cumulative environmental impacts, cellAg production of chicken, pork, and beef will reduce these by 17%, 92%, and 52%, respectively[28]. Ambitious modelling also suggests that 95% of the land and 78% of water currently used for agriculture can be freed and global warming could be reduced by 92%[29]. Approximately, 70 billion terrestrial farm animals are raised and killed for food every year, a figure that could be reduced upon transition to cellAg[24].

The so-called carbon miles of food seriously impact the emissions cost of food when transportation costs are added, accounting for a total of 41% of the global food emissions[30]. Another area that could be improved through cellAg is the production of food closer to major population centres, reducing the need for long-distance transportation. Overall, cellAg is resolving some of the most pressing environmental problems such as deforestation, loss of biodiversity, global warming, and pollution. If we combine the potential environmental, social, and health impacts of cellAg, the picture of a sustainable way for growing meat is beginning to emerge. Environmental impacts are extensively explained elsewhere in this book.

3.3 Economy

When the first cultured meat patty was developed and presented in 2013, the cost of production was more than US$300 000[31]. A variety of companies have begun to reduce these costs and to devise a way of scaling up reactor systems and recycling culture media for affordable large-scale production[32,33]. Obtaining conventional meat from an animal takes considerable time to raise the animal for slaughter. There is little room for cost reduction and improvement of feedstock efficiency as the model is governed by the growth of the animal. The cultured meat model allows for improvements in efficiency by directly producing muscle tissue for consumption. Significant cost reduction will occur with the removal of fetal bovine serum (FBS) from culture media with several companies already targeting serum-free media products[34–36]. Cultured meat allows for products that would otherwise not be available in the market, for example exotic meat sources or seafood in inland communities. It is hoped that countries with little land area for cattle production, which are dependent on imports, could produce cultured meat. Furthermore, compared to conventional meat, cellAg could allow more people across the globe to access high-quality meat at a cost favourable both economically and to the environment[2,3].

4 Advances in cellular agriculture

One aim of developing cellAg technology is to deliver a meat product that is safe, nutritious, sustainable, and that can meet the demand for food, which

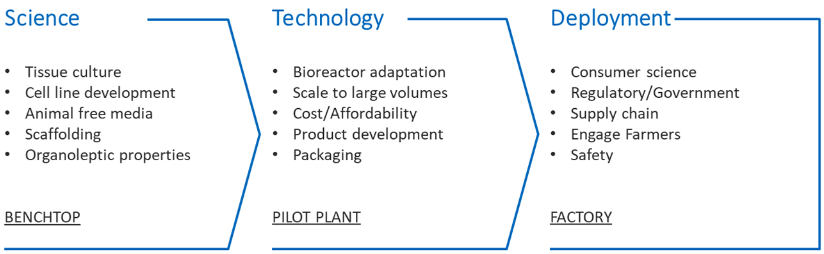

Figure 4 Technology suite and key development requirements for the successful launch of cellAg products in the market. Innovation and discovery at the laboratory scale continue to be required for the discovery and development of alternate solutions. Conceptually, in a subsequent workflow, technology is required to scale and adapt laboratory procedures to pilot plant and factory scale. Consumer and market studies are required to launch and deploy safe consumer-grade products in the context of a defined regulatory framework.

traditional livestock is increasingly struggling to provide[37]. As this is a novel field, new advancements are continually being developed by a wide range of stakeholders including both academic and private research laboratories. The field is based upon multidisciplinary research in the biological sciences, engineering, and agriculture. Key areas for development include cell line development, production of cost-effective cell media, development of cell scaffolds, product development, and engineering of novel bioreactor systems to produce quantities of biomass required at a commercial level (Fig. 4).

4.1 Discovery at laboratory scale

Cells and cell line development are at the initial core of cultured meat development. While some groups are interested in the development of liver tissue from hepatocytes or fat tissue from adipocytes, most edible meat originates from muscle tissue and therefore most R&D efforts are focused on muscle cells. Currently, the main cell line used for fundamental science and research in the muscle research field is the C2C12 myoblast cell line derived from mice. Another publicly available myoblast cell line is derived from quail (QM7). The growing cellAg field is pushing for the generation of further myoblast lines derived from commonly farmed livestock animals. The two main types of stem cells utilized are pluripotent stem cells and adult stem cells. Muscle satellite cells, mesenchymal stem cells (MSCs), and fibro (adipogenic) progenitors (FAPs) are the three main adult stem cell lines[38]. Embryonic (ESCs) and induced pluripotent stem cells (iPSCs) are the two main pluripotent stem cell line types developed. Recently, the Good Food Institute funded projects to develop cell lines from cows, pigs, shrimps, and fish. Research on their viability and myogenic potential is under way[33].

The identity of a cell usually determines its physiological behaviour. Most eukaryotic cells display an adherent phenotype, i.e. their growth and development are dependent upon being attached to a surface. For growth at scale in bioreactor vessels, this can be unfavourable and cells growing in suspension would be preferred. Interestingly, for the application of CellAg, a combination of both properties might be most desirable. In this way proliferation to high cell density might work better in suspension, whereas differentiation into edible biomass might work better with adherent cells. At this point, there is no consensus on how this impasse can be resolved best, and there have been advances in turning other cell lines like CHO into suspension[39]. Different forms of microcarriers are being explored for their potential to provide surface and buoyancy so that a bioreactor can be filled in three dimensions with growing biomass. Moreover, by using cellular aggregates, novel approaches are being explored to overcome this dilemma[40].

For decades, cell and tissue culture work has been utilizing foetal bovine serum (FBS) in conjunction with a basal medium consisting of nutrients and salts required for cell growth. In the context of cellAg, FBS is unfavourable for several reasons. It is expensive, it is animal derived, and, based upon its origin, it varies in composition and quality. Another compelling thought is the idea that FBS will become less available if cultured meat is as successful as anticipated. At present, in the cellAg field, few FBS-free media formulations are available including Essential 8[41] and TeSR[42] medium. Research is being conducted to create chemically defined media, often utilizing recombinant growth factors, which efficiently can replace serum-based medium and animal-derived growth factors. A variety of strategies are also being employed including cost reduction of recombinant growth factors or minimizing the concentrations required[35].

Preparation and optimization of cell media is the single most crucial step to developing a meaningful supply chain for the production of cultured meat. Both academic and corporate institutions drive media development such as Mosa Meat B.V., one of the early start-up companies, which managed to reduce their medium cost by 98%. This was achieved by characterizing myogenic gene expression during differentiation and by supplementing the corresponding regulators in the media to enhance its definition[36]. Other companies such as UPSIDE Foods[43] and CellMEAT[44] have claimed the development of FBS-free media as well. A range of media formulations have been developed by academic institutions, such as Tufts University developing serum-free medium for bovine cell lines[35] and differentiation media for rapid muscle cell differentiation. At present, about two dozen cell media suppliers are ready to cater their products to the cultured meat industry[33].

Animal-based meat takes shape on bones with intercalation of fatty layers and collagen-based connective tissue. Without these features muscle tissue would be pointless, and it is their properties which impart texture and

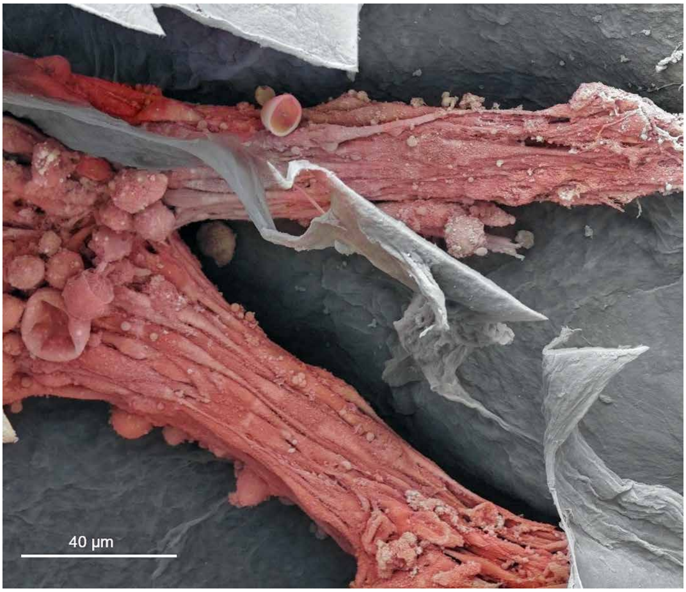

Figure 5 Scaffolding at the cellular level.[45] SEM image of differentiated myotubes (C2C12 cells) bound to a nanocellulose bioscaffold (NBS) matrix. Understanding cell-to-matrix interaction will be helpful for the successful scaling of cultured meat.

mouthfeel to a piece of animal-based meat. To create three-dimensionality, bite, texture, and mouthfeel with cultured meat, various approaches are targeted towards the development of scaffolding materials, which are combined with the growing cell mass. Suitable scaffolding is also needed for adherent muscle cells to enable a structure, which ensures efficient nutrient and waste movement to maximize the growth of biomass. Ideally, scaffolding material would be edible and potentially add nutrition to any final meat product[45]. Several types of scaffolds have been successfully developed, such as hydrogel-based, algae-based scaffolds, and nanocellulose[38] (Fig. 5).

4.2 Technology development and scaling

To deliver cultured meat products into regional and global markets, sizeable amounts need to be produced and current concepts pursue the development of bioreactors at various types and levels. In principle, a bioreactor is a vessel that provides controlled conditions regarding temperature, oxygenation, pH value, medium quality, and physical agitation. The best bioreactors – by far – are live animals. Animals provide structural support through bones, cartilage, and collagen to grow muscle tissue. Moreover, a live animal organism is equipped with blood vessels to provide for the transport of nutrients and cellular waste products. Researchers are trying to replicate the conditions for cell growth

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found in animals to accommodate the growth of animal cells outside their body. Credible scepticism has been expressed towards the idea of growing 'meat' in bioreactors at volumes comparable to volumes delivered by the livestock industry[46]. More on techno-economic analysis will be discussed elsewhere in this book.

Based upon the cellular system and the product of choice, i.e. muscle meat, foie gras, or seafood, etc., the size, process, and design of the bioreactor are selected. Conventional bioreactor systems fall into various categories such as stainless-steel stir tank bioreactor, single-use stir tank bioreactor, orbitally or rotationally shaken, fluidized bed, rocking wave, or hollow fibre bioreactor (HFB). Additional options include airlift and bubble column, vertical wheel, and fixed bed bioreactors. Advances have been made using several types of bioreactors for C2C12 cell proliferation, such as HFB[47] and polysulfone hollow fibres[48]. Currently, bioreactors of up to several thousand litres volume are being utilized in the cultured meat field[49]. As it has been established for the biomedical industry, for every run of a bioreactor, an optimized seed train needs to be developed consisting of several sequential steps to expand an aliquot of frozen cryoculture via shaker flasks and intermediate culture devices to the desired final main culture.

Regardless of the achievements made in scaling up of cultured meat, over the foreseeable time, pure cultured material might not be competitive in price to animal-derived meat. Therefore, with the idea to provide high-quality blends of nutrient-dense cultured meat together with fibre-rich complement, many developers pursue hybrid products together with plant-based material. Both plant and cultured approaches have their benefits and can be combined to create a range of products that cater to a spectrum of consumers. Creating a product from both will have the advantages of being nutritious, clean, and sensory equivalent to conventional meat. Interestingly, the difference in overall content at the level of the final product between a bio-scaffold used for growth of the cells and subsequently added plant material to produce a hybrid product could be exceedingly small (Fig. 6).

4.3 The emerging cellular agriculture industry

After the first 'proof of concept' hamburger had been developed and presented in 2013, the number of innovators in the new research field saw significant growth year after year. Witnessing Prof. Mark Post serve a cultured meat hamburger at a media event in London[50] has triggered a chain of events resulting in multiple cultured meat start-up companies. Currently, the number of start-ups focused on developing cultured meat products has risen to more than 100, with approximately 21 new cultured meat ventures compared to the previous year[33].

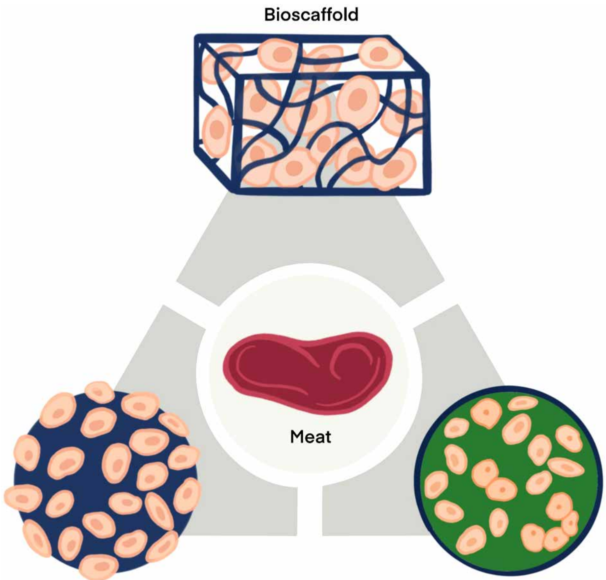

Figure 6 Successful scaling necessitates firm control over the interaction between myoblast cells and their environment at various levels of magnitude and at various time points during the production of cultured meat. At the micrometer level, it will be desirable to achieve a high ratio of cells per microcarrier. Bioscaffolds are being employed to impart dimensionality and texture to the growing biomass. Many current approaches in the emerging cultured meat industry are boosting production levels by the introduction of combined materials using cultured and plant-based materials. Thus, the edible product indicated in the centre might range from a blend of cultured meat with a high plant content to a meat without any plant material. Ongoing technology developments will provide for more seamless transitions between the various levels of interaction.

The cultured meat field has grown exceptionally and has managed to reach an investment capital of $1.3 billion in 2021[33]. Cultured meat companies such as Believer Meats (formerly Future Meat Technologies), GOOD Meat (formerly Eat Just), and UPSIDE Foods (formerly Memphis Meats) have shown evidence as to how far this field has advanced in just 8 years. To enable rollout in the market, the Singapore food authority (SFA) issued regulatory approval for the sale of cultured chicken bites by GOOD Meat[51].

CellAg aims at utility broader than replacing traditional meat products with the declared interest in other animal-sourced goods. Animal-derived products used in fashion or cosmetic industries represent another potential area for research and development, e.g. cultivating gelatine, fur, and leather[52]. Other animal food products such as milk[53], eggs[54], and caviar could also be replicated by cellAg advances. One example to note is the production of cultured human breast milk, which could represent an interesting alternative to infant formula[53].

Some companies expressed interest in using cultured meat products in the pet-food industry. Similarly, biomedical research and the biomedical industry may also benefit from the advances of cellAg with FBS representing a significant cost in cell cultivation for a range of products such as antibodies and vaccines. Growth media formulations without FBS may help to minimize any batch effects introduced by the addition of FBS.

5 Challenges for the cellular agriculture field

Cultured meat is based on the idea of producing meat and other animal products without animals. While this field has the potential to revolutionize the way we think about food, it also needs significant development and technological innovation. As mentioned, the challenge lies in replicating the muscle-growing environment of an animal inside a technology platform or a production site. Our knowledge of tissue engineering and development has been limited to medical applications and research[55]. Overall, the biggest challenge for the cellAg field is scaling at affordable cost. This challenge can be addressed by developing solutions to the speed of cell growth, to the manageable volume of cells that can be grown safely in a bioreactor, and to the details related to the procurement and production of cell media. Existing technologies will have to be altered to be applicable at a scaled-up stage. Discussed below are some of the technical challenges.

5.1 Cell line procurement and media optimization

One challenge in developing cultured meat involves maintaining the viability of cells *in vitro* and the ability to both proliferate and differentiate into the various cell types and structures[56]. Well-developed cell lines sourced from commonly consumed animals are important to cellAg as they directly impact the downstream upscaling, and therefore, the end-product[57]. Induced pluripotent stem cells (iPSCs) have been developed to mitigate the complexities of deriving cells from an embryo; however, they possess properties of embryonic stem cells[58]. Overall, it will be important to obtain better control over the differentiation/proliferation balance for different cell types used[56].

As mentioned before, FBS is a key element of traditional cell culture supplementing the media with essential nutrients, proteins, and fats. FBS is derived from the blood of a dairy cow foetus when it is being slaughtered. In addition to ethical concerns, there is a limited supply, leading to a significant price. Using serum would drive up the cost of cultured meat as FBS can cost upwards of $500 per litre[59], and approximately 50 litres are required to culture one kilogram of meat using current conditions[46]. Moreover, FBS is undefined, which can complicate the scale-up process and hamper the health of the cells at a commercial level[60]. To make cultured meat 'clean' and sustainable, it needs to be free of FBS. Thus, a renewable and accessible alternative is required that can be scaled up to accommodate the new industry.

5.2 Bioreactor adaptation

Another obstacle preventing cellAg from easily being adopted commercially is the inability to produce sufficient biomass[61]. In part this obstacle is based upon problems associated with industry-sized bioreactor design. Imperfect modelling, mass transfer, and shear stress are some of the limitations being faced[62]. Currently, modelling techniques for large-scale bioreactors are ineffective, with smaller-scale bioreactors unable to predict conditions for large-scale bioreactors[61]. Moreover, Computational Fluid Dynamics (CFD) tools used to model the operation of large-scale bioreactors are not configured for a cellAg context and are thus unreliable[63]. In lab-scale cell culture vessels, the large surface area to volume ratio means that the oxygen mass transfer occurs through the surface of the media. However, large-scale bioreactors have a much smaller surface area to volume ratio and rely on sparging and mixing as the main mechanism of oxygen mass transfer. Thus, the ability to predict and control the rate of oxygen transfer through sparging becomes a crucial design challenge in upscaling bioreactors.

Ongoing innovation and research are being dedicated to the reduction of shear stress in the bioreactors, which refers to the force experienced by cells inside the bioreactor because of velocity gradients present in the fluid following mixing or sparging[64]. The amount of shear stress is a significant factor in bioreactor design since it can cause cell death or detachment of anchorage-dependent cells from microcarriers (MCs)[62] or from bioscaffolds.

5.3 Scaffolding material

At present, methods to culture unstructured meat products are available, but not for structured products[65]. A scaffold will be required to produce structured meat products (defined meat cut analogues) such as steak. It will be easier to develop products such as minced meat[66]. Ideally, any scaffold used in cellAg would be non-animal in origin, have surface characteristics that enable cellular attachment, be affordable and easily scaled in a supply chain, and finally be able to add nutritional value to any final product.

There have been developments in using cellulose and decellularized spinach leaves as scaffolds that allow cells to grow in a defined arrangement[67,68]. This is currently on a small scale and will be required to be scaled up to produce meat in large quantities. Using 3D printing tools to impart structure and texture is being considered as an alternative, although that would add up to the overall cost of the final product[69,70].

5.4 Acceptance and marketing challenges

Numerous challenges are associated with cellAg technology unrelated to scaling and the product development areas of media, bioreactor, and cost optimization. Of equal importance is the ability to market, at scale, cultured meat products, hence the significant interest in consumer expectations around cellAg products.

Even though multiple scientific publications and reports have been published around consumer expectations in the cellAg domain, there is currently no market for cultured meat[71]. Consequently, demand cannot be precisely assessed, and only forecasted.

Consumers have clear expectations towards the organoleptic properties of meat[14]. To achieve these properties with cellAg will be a challenge for everyone in this new field. It seems clear that a consumer who wants to eat meat will eat meat. Some of the published literature suggests that the proportion of participants willing to try cultured meat varies significantly, from 5% to 11%[26]. A key factor that reduces the acceptance of cultured meat is the belief that 'what is natural is good'[72]. Public and food-related risk awareness among individuals also influences the decision to accept or reject cultured meat[73]. These factors, whichever way they are mixed among populations, will affect the overall success of cultured meat products.

6 Conclusion and future trends in research

Many of our twenty-first-century food systems are suffering from parallel crises, i.e. climate change, sociocultural conflicts, large-scale global interdependencies, pandemics, and limited biodiversity – to name the most visible. CellAg and the various cultured products are being associated with numerous advantages ranging from being more sustainable, being more ethical, being nutritionally superior, to being more convenient and one day even more affordable. It will, however, require remarkable development and scientific progress to establish a robust system that can support the demand for meat. If the challenges linked with technology development, economic implementation, and consumer acceptance are being met, it is reasonable to assume that the expected advantages indeed will materialize.

For the future, it is realistic to anticipate different product offerings in the meat segment for consumers. Animal-based meat products will be co-existing with plant-based and cultured meat. It seems unrealistic to anticipate that animal-based products will be disappearing from the supermarket shelf anytime soon. Over the long run, it will be desirable to observe an alleviation of issues related to animal and livestock handling and to practices linked with factory farming.

The enthusiasm behind novel technology development is credible, and it is important to push financing, research, and implementation to the next level. At the same time, and as often, it is likely that the current and pioneering spirit in the cellAg field sooner or later will give way to a more sober and realistic view of the situation. A successful cellAg industry will have the visual appeal of stainless-steel bioreactor farms in an ultraclean production environment. Throughout its production, cultivated meat will raise the bar for hygiene and food safety as compared to conventional animal-based meat.

7 Where to look for further information

7.1 Introductory resources

- 'How it's made : the science behind cultured, clean, and cell-based meat' by Elliot Swartz (https://elliot-swartz.squarespace.com/cellbasedmeat/cleanmeat301).
- 'Is the future of meat animal-free?' by Liz Specht (https://www.ift.org/news-and-publications/food-technology-magazine/issues/2018/january/features/cultured-clean-meat).
- 'Cultured Meat and Future Food' podcast by Alex Shirazi (https://podcasters.spotify.com/pod/show/futurefoodshow).

7.2 Further reading

- Good Food Institute cultivated meat publications list (https://paperpile.com/shared/WLg4bc).
- The New Harvest OpenCellAg Repository (The New Harvest OpenCellAg Repository | Zenodo).
- Good Food Institute 'State of the industry reports' (https://gfi.org/resource/cultivated-meat-eggs-and-dairy-state-of-the-industry-report/).
- 'FDA completes first pre-market consultation for human food made using animal cell culture technology' (https://www.fda.gov/food/cfsan-constituent-updates/fda-completes-first-pre-market-consultation-human-food-made-using-animal-cell-culture-technology).
- 'Ex-ante life cycle assessment of commercial-scale cultivated meat production in 2030' (https://link.springer.com/article/10.1007/s11367-022-02128-8).

7.3 Books

- 'An introduction to cellular agriculture' by Ahmed Khan.
- 'Moo's Law: An investor's guide to the new agrarian revolution' by Jim Mellon.

- 'Clean Meat: How growing meat without animals will revolutionize dinner and the world' By Paul Shapiro.
- 'Billion Dollar Burger: Inside Big Tech's Race for the Future of Food' by Chase Purdy.

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