

Advances in the Development of Meat Analogues and its Role in Prevention of Nutritional Deficiency in Vegetarians - A Review

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Abstract: Meat is regarded as the best quality protein source not only because of its nutritional qualities but also because of its valued flavor. Meat is also important in terms of sustainability since it is one of the most energy-intensive and environmentally damaging food. As part of their eating habits, the worldwide population is on the lookout for nutritional and environmentally beneficial supplies. While animal products are high in protein, the appropriateness of vegetarian protein intake has long been a point of contention. While the lower protein intake and quality of protein in a vegetarian diet are commonly thought to be a concern, there is growing evidence that eating protein from plants rather than animals may be one of the reasons why vegetarians have a lower risk of being overweight, obese, or suffering from chronic disease. Plant-based meat substitutes that closely resemble the structure and feel of meat are gaining popularity. Meat analogues or plant-based products that mimic the characteristics of typical meat products have established a place in the protein food discourse. Veganism popularity, along with growing concerns about animal welfare, detrimental effects on human health and the environment has fueled demand for meat replacements, mostly plant-based meat analogues.

Keywords: meat analogue, protein sources, vegetarian, plant based product, nutrition

1. Introduction

Meat is one of the most important sources of animal protein in the diet. It is a vital part of a healthy diet since it contains critical nutrients such as protein, lipids, vitamins and minerals (Pereira and Vicente 2013). During the projection period 2020–2025, the global meat market is expected to develop at a compound annual growth rate of 7.35 percent (Boukid 2020). At a compound annual growth rate (CAGR) of 16 percent, the global cultured meat market is predicted to expand from \$110.09 million in 2020 to \$127.67 million in 2021 (2021 cultured meat market). Despite its long history of use, campaigners and government agencies are mounting pressure on meat producers to cut production due to ethical and environmental concerns. This is because the studies show that raising cattle for protein harvesting has a profoundly bigger environmental impact than producing the same amount of protein from non-meat sources like grains, beans and seeds (Nijdam et al., 2012). Livestock animals are raised for meat production, which necessitates the use of extensive amounts of land and water, resulting in increased greenhouse gas emissions and a negative impact on the environment (Gerber et al., 2010). Foodborne viruses are common in meats and responsible for various diseases. Furthermore, red meat consumption can lead to ischemic heart disease, exacerbate the obesity pandemic and raise the risk of joint inflammation and colon cancer (Larsson & Wolk, 2006). If we keep our current per capita protein consumption, the world's population will grow, necessitating more edible protein production (Jones, 2015). As a result, more people choose to eat less meat or eliminating it entirely from their diets for health reasons (Sun et al., 2020). Alternatives to conventional animal products are being developed to lessen the environmental challenges, relieve public health issues and make food system more stable (Sun et al., 2020). A meat analogue is a food product that closely

resembles the appearance and chemical properties of specific types of meat. These are made from non-animal protein and have a similar appearance and flavour of meat. Cost advantages, relatively stable price due to decreased proneness to seasonal supply swings, a longer shelf life and easier storage make such foods appealing to the processing sector (Kumar et al., 2017). Vegetarians, vegans and non-vegetarians who desire to reduce their meat intake for health or ethical reasons, as well as people who follow religious dietary guidelines such as Halal, Kashrut, or Buddhist, are all part of the meat imitation market. Some vegetarian meat substitutes are manufactured using centuries-old recipes for wheat gluten, rice, mushrooms, legumes, tempeh, or pressed-tofu, which are then flavored to taste like chicken, beef, lamb, ham, sausage, shellfish and so on (Malav et al., 2015). Textured vegetable protein (TVP), a dry bulk commodity generated from soy, soy concentrate, mycoprotein-based Quorn (which utilizes egg white as a binder, making it unfitting for vegetarians) and modified defatted peanut flour are some of the more recent meat analogues (Malav et al., 2015).

Prime nutritional function of meat in the diet is to provide high-quality protein. If meat is completely swapped by meat-analogue products, the nutritional content of those items should be comparable. It has been found that a meat analogue with a protein content of up to 30 percent and a low fat and lipid content can be a viable nutritional substitute to meat (Kyriakopoulou et al., 2019). The criteria are less stringent when meat analogues are used to partially replace meat in a diet. Stronger substitution of meat with food of different composition might aid in achieving a more balanced diet (Wild, 2016). To put it another way, reducing meat consumption may offer extra health benefits (Kyriakopoulou et al., 2019). Meat substitutes do not include saturated fatty acids, cholesterol, or purines and hence do

not have the negative health repercussions associated with excessive red meat consumption. Furthermore, low-meat and low-dairy diets contain a high dietary fibre content and numerous phytonutrients that promote health (Craig, 2010). Plant based diets have been shown to provide a variety of health related advantages that includes prevention of obesity, induced metabolic dysfunction, cardiovascular disease and diabetes other than providing anti-cancerous and anti-inflammatory function (Zhang et al., 2016). Further, the strong satiety effect and higher expenditure of plant proteins may contribute to weight reduction and weight maintenance (Kristensen et al., 2016).

This review therefore aims to delve into the advancements in the development of the meat analogue over the years. This article looks through the developments in the various aspects of the meat analogue in terms of its nutrition, benefits, market, consumer demand, composition etc. thus, giving a good idea about the importance and urgency of shifting to a plant based diet for the sake of our health and the environment.

Advances in the development of meat analogue

The idea of providing a more sustainable alternative to meat-based diet through the implementation of meat analogues has brought a significant change in the food landscape of the world. Meat analogues or meat simulation products are one of the few products where consumers are giving priorities over the purchase and the consumption of ultra-processed foods (Bohrer, 2019). Awareness to safer and more environmentally friendly food consumption has steered many people toward a plant protein-based meat substitute, which necessitates that the new products meet market acceptance expectations by making a significant contribution to resolving current environmental challenges (Fonmboh et al., 2021). The replacement of plant-based products for meat in part or whole is seen as an evolving strategy for reducing excessive consumption of meat and animal protein products. Plant proteins are therefore a versatile source of animal protein and meat substitutes are one of the most effective ways to incorporate plant proteins into human diets in order to provide a broader variety of protein options (Fonmboh et al., 2021).

1.1 History of meat analogues

The development of meat analogues was not a fast and spontaneous one. It is a subject that has its own history, which went through different iterations and different looks at different times. It had different variations and went by different names. Plant-based meat substitutes aren't a fresh food category and they aren't even a novel idea. Several previous studies also covered the long history of meat analogues and other plant-based meat substitutes that mimic the characteristics of meat. Most importantly, Shurtleff and Aoyagi (2014) published a timeline outlining the evolution of meat alternatives from 965 CE to 2014 with hundreds of chronological events happening all over the world. The earliest known reference to a soy-based tofu product was in China in 965 CE. According to this article, many of today's meat analogue manufacturing innovations such as biopolymer spinning and extrusion were first patented in 1947 and 1954 respectively. Tofu (a soy

product) dates back to 965 CE, wheat protein to 1301, Yuba to 1587, tempeh to 1815 and combinations of nuts, cereals and legumes to 1895 (Shurtleff et al., 2014).

A global expansion of the modern meat analogue marketplace started to occur in recent years (2015 to 2020) with rapid growth in product offering and availability. As said by Kyriakopoulou, "the marketplace for meat analogue products in Europe and North America has expanded beyond just vegetarian consumers to now include meat eating and meat loving consumers." (Kyriakopoulou et al., 2019). While historically structured products such as East Asian tofu, seitan and tempeh as well as African Chikanda, Kinaka, Napsie and Nyam ngub have been around for decades, meat analogues research has only been around since the early 1960s in Europe and the Americas. Many of the new technology used to produce meat analogues today were first patented between 1947 and 1954. As a result, the limited understanding of meat analogues and the shortage of individuals employed in the domain impede the creation and production of meat alternatives, especially plant-based products (Fonmboh et al., 2021).

1.2 Protein sources and composition of meat analogues

The progress in the development of meat analogues expanded to its other aspects too. From the ingredients used in the production to the extraction and the processing of the necessary proteins from non-animal sources, the meat analogues sector has undergone significant progress in development over the time. While most vegetable, grain and microbiological systems are high in protein, differences in the physicochemical properties of those proteins have made developing optimal extraction and processing techniques for meat-analogue products difficult. Protein extraction methods have improved in recent years, as has innovative processing to aid physical structuring and the detection of protein extracts with superior physical properties for meat-analogues. Improved extractions and processing of proteins from conventional (e.g. soybean) and atypical (e.g. chickpea) sources have been the subject of recent developments in non-animal proteins as substitute ingredients in traditional meat products (Jones, 2015).

1.2.1 Soy and cereal proteins

Soy protein has long been the most popular protein used in meat substitutes in history since long. Several observational studies have been used to compile detailed reviews on the beneficial effects of soy protein intake on lipid metabolism and cardiovascular health. Processed soy protein (i.e. isolated soy protein and soy protein concentrates) has been shown to have a higher abundance of essential amino acids than unprocessed or minimally processed soy protein in terms of nutrition (Hughes et al., 2011). Soy protein has been able to obtain a protein digestibility score which is comparable to animal derived foods like meat, eggs and dairy products (Bohrer, 2019). But recent studies have encouraged the use of additional protein sources other than soy ingredients, both for nutritional and functional reasons like as shown in Table 1. Cereal proteins are divided into many groups depending on the plant from which they came (e.g. wheat, rice, barley and oats) and the degree of processing they received (e.g. seeds, flour, isolates,

flakes).Seitan, which dates back many centuries is the most historically used form of cereal protein in meat analogue products; however, rice, barley and oat ingredients are popular among the ingredient labels of modern meat analogue products (Malav et al., 2015). This can be seen in Table 2 where a comparison of the nutrient profiles of the major meat analogues against beef is shown. From a structural standpoint, cereal proteins are extremely useful to meat analogue manufacturers. Most cereal proteins have a visco-elastic structural network which can aid in the formation of a successful bind and provide the required consistency in meat analogues while contributing to the fibrous-like texture found in ground meat products (Kumar et al., 2016). Several studies have been ongoing in order to study the effects of the addition of different ingredients on the flavor of meat analogues. Guo et al.(2020) investigated

how the inclusion of wheat gluten affected the retention of volatile taste compounds, microstructure, moisture distribution and secondary protein structures of meat analogues made by a high moisture extrusion technique. Their findings revealed that wheat gluten and moisture levels in raw wheat are linked. By altering the microstructure, water distribution, protein structure and interaction force of meat analogues material can manage the retention rate of volatile taste compounds thus allowing a wider range of goods to be developed according to various customer needs. Frazer et al.(2018) assessed the safety of the use of Soy Leghemoglobin Protein for the use as a catalyst of flavor in meat analogue production. They used both in vitro and in vivo models to assess the safety of LegH Prep in this investigation.

Table 1: Major non-meat protein sources suitable for meat analogue

S. No.	Type of protein	Source	Product	References
1.	Glycinin, Vicilin	Legumes	Burmese tofu from chickpea	Li Day 2013 Lim T.K 2012
2.	Beta-conglycinin	Soybean	TVP, Tempeh, Tofu, Kinema	Chen et al. 2009 Bohrer 2019
3.	Mycoprotein	Fusarium venenatum (Filamentous fungus)	Quorn	Elzerman et al. 2013 Finnigan 2011
4.	Gluten, Gliadins, Glutenins	Wheat, rye, barley	Seitan	Sun et al. 2020 Green and Cellier (2007)
5.	Legumin, Albumins, Globulins, Glutelins	Oil seeds	TVP	Jones 2016 Riaz 2011

Table 2: Comparison of nutrient profiles of alternatives of meat with beef

Food	Energy (kcal)	Protein (g)	Fat (g)	Starch (g)	NSPa (g)	Iron (mg)
Meat						
Beef- lean, raw	123	20.3	4.6	0	0	2.1
Soya Beans						
Tempeh- raw	166	20.7	6.4	4.6	4.3	3.6
Textured Vegetable Protein- dry	257	51.5	0.8	4.2	21.0	-
Wheat Protein						
“Tivalli”- Burger, raw	127	17.0	5.0	1.5	5.0	2.1
Pea Protein and Wheat Protein						
Arrum- Dry	345	26.0	1.4	-	2.5	-
Myco- protein						
Quorn- raw	86	11.8	3.5	Trace	4.8	-

Table sourced from (Davies and Lightowler 1998)

12.2. Legume proteins

Legume proteins (pea, lentil, lupine, chickpea, mung bean and others) have become increasingly common among meat analogue manufacturers in recent years. The promising application of pea protein when structured with high-moisture extrusion was explored by Kyriakopoulou et al. (2019). Legume proteins have many promising and special processing characteristics thus making them an excellent alternative to other protein ingredients (Bohrer, 2019). Pea protein, which has been structured by high-moisture extrusion, is the most promising for meat-analogue applications. Pea-based structures, on the other hand, are softer than soy-based materials. Studies are being conducted to improve the gel strength by modifying protein hydrogen bonding such as by adding chaotropic ions to salts or by optimizing processing conditions such as temperature, protein particle size and so on. Studies have demonstrated strong emulsion and foam stabilization properties of chickpea, lentils and lupine (Sun & Arntfield, 2012).

From a functional standpoint, legume proteins complement other protein ingredients well and have a variety of promising and special processing properties. Meat does not contain carbs unless it has been further processed and carbohydrate components have been added, which is frequent in meat processing particularly in emulsified and formed processed meat products. Carbohydrates are nearly always present in meat analogue products. Carbohydrates can originate from a range of sources in meat analogue products and those sources can serve a variety of functions in the production process (Bohrer, 2019). A comparison of the nutritional composition of meat products and meat analogues is shown in Table 3. Carbohydrates can be beneficial to health in the form of more dietary fiber or harmful in the form of more refined starches or sugars from a nutritional viewpoint. Both meat analogues and processed meat products often contain a combination of dietary fiber, carbohydrates and sugars in their composition. Therefore, we can conclude that it is an added health advantage when it

comes to the carbohydrate components used in meat analogues (Bohrer, 2019).

Table 3: Nutritional composition of meat and meat analogue products

Product	Energy value (kcal)	Protein (g)	Fat (g)	Saturated fat (g)	Cholesterol (mg)	Total carbohydrates (g)	Dietary fiber (g)	Na (mg)	Fe (mg)
Meat Analogue Product									
Beyond Burger	221.24	17.70	15.93	5.31	0.00	2.65	1.77	345.13	3.72
Impossible Burger	212.39	16.81	12.39	7.08	0.00	7.96	2.65	327.43	3.72
Morning Star farms grillers original burger	203.13	16.81	7.81	0.78	0.00	12.50	6.25	609.38	1.72
Boca all American veggie burger	140.85	18.31	5.63	1.41	7.04	8.45	5.63	492.96	2.39
Garden meatless meatballs	166.67	15.56	7.78	0.56	0.00	10.00	3.33	355.56	8.33
Tofurky ham roast with glaze	203.70	20.37	5.56	0.46	0.00	18.52	0.93	592.59	1.76
Quorn brand chicken nuggets	203.39	10.17	8.47	0.42	6.78	24.58	5.93	449.15	0.72
Traditional meat products									
Ground beef (93% lean, 7% fat), uncooked/raw	152.00	20.85	7.00	2.89	63.00	0.00	0.00	66.00	2.33
Ground beef (93% lean, 7% fat), cooked, pan-fried	182.00	25.56	8.01	3.29	84.00	0.00	0.00	72.00	2.82
McDonald's beef patty	266.67	23.33	20.00	8.33	83.33	0.00	0.00	400.00	3.33
Tyson fully cooked home style beef meatballs	300.00	15.56	16.47	5.88	47.06	5.88	1.18	352.94	2.12
Hormel cure 81 classic boneless ham	105.95	18.45	3.57	1.19	50.95	0.24	0.00	1038.10	0.83
Tyson fully cooked chicken nuggets	300.00	15.56	18.89	4.44	44.44	16.67	0.00	522.22	0.91

Table sourced from Fohmboh et al. 2021, Bohrer 2019

1.2.3. Mycoproteins

Another development we can observe in meat analogues is the addition of mycoproteins as an eco-friendly protein alternative. Mycoprotein is a fungus-derived protein that was first identified as an environment-friendly protein substitute in the 1960s. Mycoprotein is made using a variety of fermentation and processing methods and several studies have looked at its use in a variety of food applications (Finnigan, 2011). Dietary fiber is contained in hyphae cell walls. Polyunsaturated fat is found in cell membranes, while high-quality protein is found in the cytoplasm. It's cholesterol-free and low in saturated fats, with a good fatty acid profile and fiber quality that rivals other vegetarian protein sources. Consumption of mycoprotein lowers blood cholesterol levels dramatically and can promote weight loss due to its high fiber content. In general, the filamentous fungus is preferred for the development of meat substitutes because it is thought that the mycelia will give the final product a fibrous texture similar to that of meat. Natural mycoprotein, in the form of mushroom mycelia, has been used which has fibrous structures that give a distinct chewing sensation. The edible filamentous fungus *Fusarium graminearum* was used to create the first commercial meat analogues, such as burger patties and sausages with mycelia, which passed the UK Food Standard Committee's safety examination in 1994. Because these meat mimics have high sulphur content amino acids and glutamic acid, they naturally have a meaty flavour indicating the umami flavor (Kim et al., 2011).

Traditional meat analogues have a low lipid content; however, modern meat analogue products have a far higher lipid content than traditional meat analogue products. In reality, modern meat analogue products have approximately the same lipid content as traditional meat products. In the same way as a variety of lipid ingredients (fats/oils) are used in the formulation of meat analogues. Several research studies have been used to compile comprehensive reviews

on the beneficial benefits of soy protein consumption on lipid metabolism and cardiovascular health (Bohrer, 2019). Canola (rapeseed) oil, coconut oil, sunflower oil, corn oil, sesame oil, cocoa butter and other vegetable and plant oils are among the lipid ingredients used in modern meat analogues (Bohrer, 2019). As stated in a previous analysis by Kyriakopoulou et al. (2019), the juiciness, softness, mouthfeel and flavor release of meat analogue compositions are aided by fats and oils (Kyriakopoulou et al., 2019). Recent developments of new meat analogues have also started the addition of cocoa butter in the most recent recipes (Bohrer, 2019). To heighten the flavour and improve texture and mouth feel, lipids rich in saturated fatty acids (e.g. coconut oil and cocoa butter) or unsaturated fatty acids (e.g. sunflower oil, canola oil, sesame oil and avocado oil) are utilized. To replicate meat flavour associated with lipid oxidation and volatiles created by the Maillard reaction during heat processing, the supply and content of fatty acids is critical (Simon et al., 2019). Vegetable oils are cholesterol-free and hence considered healthier than animal fats from a nutritional standpoint; but to achieve meat-like sensory qualities, greater attention must be made to the balance of unsaturated and saturated fatty acids. Oleogels could be a promising technique for replacing saturated fats in plant-based meat substitutes. Because of the water-binding ability of beta-glucan, fiber-rich components such as oat-hull-based ingredient or oat's soluble fibre beta-glucan have been reported as fat replacers, improving the structural properties of reduced-fat products (Summo et al., 2020).

1.3 Advances in flavor of meat analogues

Another aspect that meat analogue endured a lot of development through the years is the implementation of flavor enhancers to it. Over the past one of the drawbacks that many meat analogues faced was its inability to mimic the taste, texture and juiciness of the meat (Kyriakopoulou et al., 2019). This made consumers very difficult to bring

themselves to the consumption of meat analogues. But nowadays, seasonings and spices, much as in most processed and packaged foods are used to add specific flavor ingredients to meat analogue products. A variety of techniques have been explored to achieve the "meat-like" taste in meat analogues (Bohrer, 2019). The color of meat products is frequently regarded as the most essential factor in customer decision-making. The color of meat products varies while cooking, in addition to the color of raw meat products (mainly bright red for beef, red pink for pork, blue white to yellow for poultry), since the proteins responsible for the color of meat (mainly myoglobin) undergo chemical changes. For meat analogue goods, a similar notion may be offered for the relevance of color and color change while cooking. Before, during and after cooking, meat analogue goods should exhibit color characteristics that are comparable to the meat product they are mimicking. The substances used to produce the distinctive hues of current meat analogue products differ from one product to the next (Bohrer, 2019). The overall idea is to use components that elicit naturally occurring color characteristics that are equivalent to the mimicked meat product. This is the case with the Beyond burger which utilizes beet juice extract and the Morningstar Farms burger which uses tomato paste. Another method for achieving color characteristics comparable to conventional meat products is to employ sarcoplasmic proteins which have a chemical structure similar to the iron and oxygen-binding protein present in muscle tissue myoglobin. The Gardein vegetarian meat balls include a decreased iron molecule, but the Impossible burger contains soy leg hemoglobin, a soy-derived substance having chemical and structural similarities to hemoglobin and myoglobin, the former being a key protein responsible for flesh color (Robinson, 2019). Furthermore, soy-based meat analogues must contend with customer concerns about soybean genetic modification (GM). Despite the fact that GM technology for changing the physical and nutritional qualities of plant proteins has progressed significantly over the last two decades, some consumers continue to be skeptical of the potential harm to human health. Heme, an iron-containing molecule, is prevalent in animal muscles and gives real meat its colour and flavor (Sun et al., 2020). Heme is created by genetically engineering yeast, by introducing the soy leghemoglobin gene to a yeast strain, fermenting the yeast and separating heme from the yeast as a crucial element in plant protein-based meat mimic products like the Impossible Burger. The addition of plant-based heme to meat substitutes can improve their robust meaty flavour, fragrance and cooking qualities. However, additional testing is required to ensure its safety (Sun et al., 2020). Impossible Foods (USA) is another notable example, with the motto "Plants + Science = Meat." Many investors have contributed to the company's funding, including Khosla Ventures, Horizons Ventures, Open Philanthropy Project and Temasek (which is supported by the Singaporean government). Their team of scientists, farmers and chefs discovered methods and ingredients that successfully mimic the appearance, scent, texture and flavour of ground beef and created the Impossible Burger as a sample meat analogue product (Sun et al., 2020). Flavor-related substances such as free amino acids, free fatty acids, nucleotides and reducing sugars are primarily responsible for the flavour of conventional meat. Aside from that, vitamin B1 and myoglobin have an impact

on meat flavour. As a result, taste enhancers are used in the production of plant-based meat analogues (Lee et al., 2020). According to Kyriakopoulou et al. (2019), a taste concentrate of meat is obtained when volatile components in conventional meat are extracted after a combination of various heat methods. Following that, many approaches for incorporating such taste concentrates into plant-based meat analogues to generate a meat flavour have been studied and developed. The addition of fat/oil can impact the flavour development, as well as the texture and mouth feel of plant-based meat analogues (Bohrer, 2019).

1.4 Processing of meat analogues

Perhaps, one of the most stark and progressive developments that occurred over the years in the history of meat analogues is the techniques of processing. The fiber spinning technique, which was developed in the 1980s was an early method for producing simulated meat. Extrusion of an alkaline protein solution into an acidic coagulating base through spinnerets resulted in filament precipitation which was then assembled into meat analogue products using binding materials. The spinning method, on the other hand, was complicated, required a highly concentrated plant protein solution and was prohibitively costly for large-scale use (Sun et al., 2020). Perhaps one of the most often used methods for the production of meat analogues can be considered as the extrusion method. The thermal plastic extrusion process has become the most common method in recent years. Extrusion processing is a well-known method for manufacturing ready-to-eat breakfast cereals and baby foods in the food industry with high productivity and energy efficiency (Liu & Hsieh, 2008). Plant proteins, typically defatted are combined with water, carbohydrates, salts, flavorings and edible lipid content before being fed into a twin-screw extruder at a high temperature and under various moisture conditions to form a meat-like fibrous framework (Guo Z et al., 2020). Several research studies related to meat analogue preparation have mentioned and practiced the use of extrusion for the preparation of their product. Chiang et al. (2018) used extrusion method to prepare meat analogues in order to study the effects of soy protein to wheat gluten ratio on its physicochemical properties. Similarly, De Angelis et al. (2020) opted the method of extrusion in order to assess the physicochemical and sensorial evaluation of meat analogues produced from dry-fractionated pea and oat proteins. Caporgno et al. (2019) extruded meat analogues based on yellow, heterotrophically cultivated *Auxenochlorella protothecoides* microalgae. Kaleda et al. (2020) opted for extrusion method to prepare meat analogue in order to study the impact of fermentation and phytase treatment of Pea-Oat protein blend on its physicochemical, sensory and nutritional properties. Vatanserver et al. (2020) reviewed about the extrusion of pulses for the preparation of plant based meat ingredients. Figure 1 shows a representation of the process of extrusion for a typical meat analogue. The advantages and disadvantages of various technologies developed recently for the production of meat analogues are mentioned in Table 4. Meat analogues are engineered to infinitely approximate the organoleptic qualities of real meat viz. texture, taste, flavor, color and sensation in the mouth by manipulating and optimizing the composition and processing parameters, in order to produce desirable goods

and increase customer acceptance (Sun et al., 2020). The new technology of 3D printing (3DP) has been lauded for its unrivalled ability to fabricate food items with complex structures while saving money and energy (Wong G et al.,2019). 3D printing, also known as fast prototyping is the process of creating solid 3D architectures from virtual models using CAD software. The CAD model is then transformed into the STL (Standard Triangular Language) file format to create digital models (Van der Linden, 2015). The application of 3D printing technology in food manufacturing has led to the development of the most important innovations in the food industry such as the creation of products with intricate shapes and complex geometries, the facilitation of supply chains and the improvement of food product accessibility (Shahbazi & Jäger, 2020). A multifaceted strategy has been proposed to counteract the detrimental effects of livestock processing which involves innovative plant materials-derived substitutes, improved waste management and policy restructurings (Ramachandraiah2021). In this regard, 3D printing (3DP) is a promising technology for fabricating customized goods with intricate shapes and textures in a long-term sustainable manner. The 3DP technology is thought to have a huge potential for lowering CO2 emissions and manufacturing product lifecycle energy demands. By the end of 2025, the global 3DP market is projected to grow to \$230–550 billion USD (Ramachandraiah, 2021). Shahbazi et al.(2021) in their research opted for 3D printing to construct a meat analogue with reduced fat levels by emulsion gels. The production of a high-quality meat analogue necessitates the use of appropriate components combined with cutting-edge technology in order to duplicate the functional aspects of genuine meat without compromising the product's characteristics. The biosurfactant-based reduced-fat meat analogues were manufactured using an extrusion-based printer to create a well-defined 3D structure. When compared to acetylated and octenyl succinic anhydride modified starches, the 3D printed meat analogues created using dodecanyl succinylated inulin and ethyl (hydroxyethyl) cellulose had a finer resolution. The findings showed that 3D printing technology may be used to create meat analogues with desired 3D structure and changed textures for improved eating experiences (Shahbazi et al., 2021).Despite the availability of several plants and byproducts as well as certain start-up ventures aiming to create food items, 3D printing of meat analogues remains

challenging. Consumer acceptance of 3D-printed meat analogues may be a major barrier to their success (Ramachandraiah, 2021).

Meat production in the lab without the need of live animals is a tremendous technical achievement made feasible by more fundamental studies into stem cell technology and muscle growth. The two most common structuring strategies for producing fibrous goods that resemble entire muscle meats are bottom-up or top-down approach to creating the fibrous morphology. Individual structural parts are generated as part of bottom-up technique and then combined into larger products. This technique is based on the structure of muscle cells, myofibrils and sarcomeres which are joined via conjunctive tissue and contain the proteins myosin and actin. This arrangement is based on the structural organization of muscle and places them at different scales from micro to macro. Culturing or biomass production of filamentous fungi, or the creation of protein fibrils or fibers are used to develop the cells. These fibrils/fibers can then be formed into a meat-like structure by cross-linking with enzymes or combining with binding agents like egg proteinand gluten (Dekkers et al., 2018).

1.5 Top-down strategy

The Top-down strategy, on the other hand, only replicates structure on longer length scales by using a force field to control structure creation or aggregation of biopolymers, resulting in anisotropic structure on a micrometer scale. This idea is used in top-down approaches like as extrusion, protein and hydrocolloid mixing, freeze structuring and shear cell technology. Shearing or freezing such mixes in a single direction can distort the scattered phase and align it in one direction. Aside from deformation, the scattered phase can also coalesce and break up, as well as inversion of the two phases, all of which affect the material's final qualities. The fibrousness in the products is caused by a continuous phase (mainly made up of proteins) that is disturbed by scattered, distorted, weak phase filaments that act as matrix/structure breakers after solidification. This has an impact on the material's mechanical properties. As a result, the structures created using these techniques are similar to the structure, but not identical to it in terms of hierarchical architecture (World Economic Forum 2019).

Table 4: Pros and cons of texturizing technologies of meat analogues

Technology	Advantages	Disadvantages	References
Extrusion	High productivity, Low cost, Versatility, Energy efficiency, Anti-nutritional factors, denaturation, Increase protein digestibility	Changes in color due to Maillard reaction, caramelization, hydrolysis and degradation of pigments	Tehrani et al. 2017 Zahari et al. 2020
High-temperature induced shearing	Cost-effective, Produce defined fibrous structure	Require more investigations for scaling	Schreuders et al. 2019 Dekkers et al. 2016
Wet-spinning	Produce defined fibrous protein produce.	Requires pure proteins -Low pH, High salt concentrations and chemical additives, Large amounts of wastes	Krinitras et al. 2015 Grabowska et al. 2014
Electrospinning	Cost-effective, Scalable, Production of very thin fibrils	Several parameters to control, Difficulties to electrospin plant proteins	Wongkanya et al. 2017 Kutzli et al. 2019
Freeze structuring	Modulation of textural properties of plant proteins	Several freezing conditions to optimize and monitor	Yuliarti et al. 2021
Mixing plant proteins and hydrocolloids	Formation of fibrous structure that can be modulated	Require hydrocolloids and the divalent metal cations for the precipitation	Mehran et al. 2013
Bioprinting (3D)	Enable the design of products with texture	Require maturation under specific	Godoi et al. 2016

printing technology)	similar to muscle fibers, Tailor the nutritional content of the product.	conditions, High production cost, The complexity of spatial structure	Voon et al. 2019
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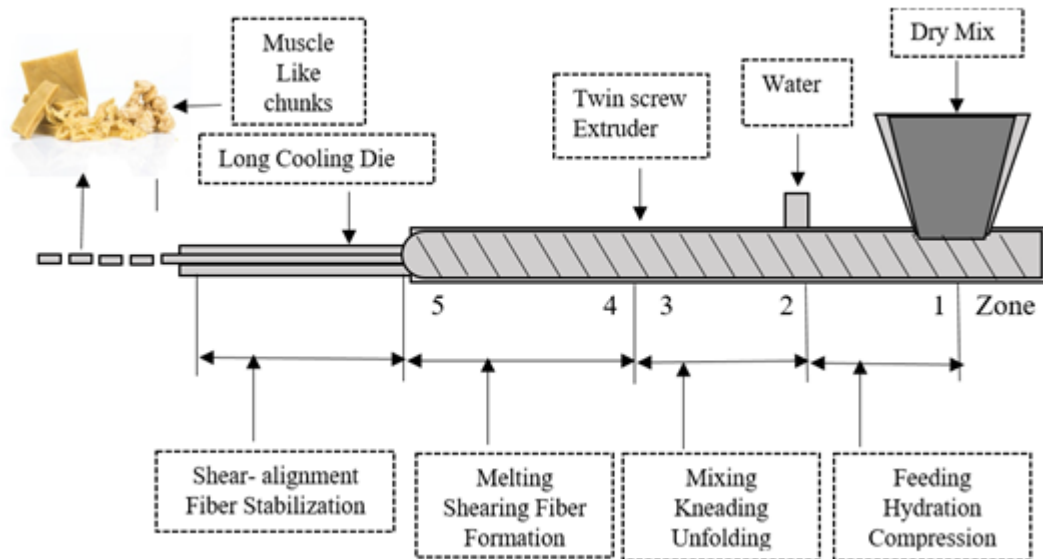


Figure 1: Process of extrusion of meat analogue

1.5.1 Wet Spinning

Wet spinning is one of the most common methods used from a long time ago to make fibrous protein products. Even though it is mostly used to create individual fibers, but it is also one of the most used methods for producing membranes for industrial separation (Girija et al., 2021). Several research have shown that food-grade fibers may be made from plant-based resources such as soy, pea and fababean. A solution containing protein is extruded through a spinneret and then immersed in a bath containing a non-solvent for solvent and non-solvent exchange culminating in the precipitation and solidification of the extruded protein phase, resulting in stretched filaments with a thickness of 20 m. The solidification mechanism determines the type of structure that is created. When the dispersion phase hardens and the continuous phase can be washed away, fibers form; when the continuous phase solidifies but the scattered phase remains liquid, capillary-filled gels form and fiber-filled gels form when both the dispersed and continuous phases solidify (Girija et al., 2021). Several research have shown that plant-based materials such as soy, pea and fababean may be used to produce food-grade fibers. The solidification mechanism determines the type of structure formed: fibers are formed when the dispersed phase solidifies and the continuous phase can be washed away. Capillary-filled gels are formed when the continuous phase solidifies but the dispersed phase remains liquid and fiber-filled gels are formed when both the dispersed and continuous phases are solidified (Dekkers et al., 2018). However, this method necessitates the use of purified proteins, a low pH, high salt concentrations and chemical additions (Liu et al., 2017). Furthermore, this procedure generates a substantial amount of waste residue, such as coagulated and washed effluent streams (Grabowska et al., 2014). MacQueen et al. (2019) used a wet-spinning approach to create beef products from gelatin fibers. Extruded gelatin microfibers mimicked the structural and biochemical characteristics of real muscle tissues during processing and cooking because gelatin is a natural component of meat.

1.5.2 Electrospinning

Electrospinning can also be utilized to make fibers for meat analogue applications (Nieuwland et al., 2014). Individual fibres of the tiniest scale are produced via electrospinning. A biopolymer solution is pushed through a hollow needle or spinneret with an electric potential relative to a ground electrode during electrospinning. Surface instabilities result from the accumulation of charge on the surface of the droplet that emerges from the spinneret, which eventually expand into very thin fibers that are attracted to the ground electrode (Schiffman & Schauer, 2008). Plant proteins are globular in their original state and following denaturation, insoluble aggregates form. The solution prepared for electrospinning proteins or protein blends with other polymers must meet a number of criteria including high solubility, viscosity, conductivity, surface tension and the ability of the components to entangle. The polymeric solution forms a Taylor cone when it is electrically attracted to a metal collector resulting in the spinning of a thread or fibril if all conditions are met. The solvent in the polymeric solution evaporates during this process, leaving thin dry strands of entangled polymers (Kyriakopoulou et al., 2019). Electrospinning produces fibers after the solvent has evaporated; thus, drying is the process of solidification. Electrospinning, according to Nieuwland et al. (2014), necessitates highly soluble protein. As a result, this technique works only in a small range of protein conductivity, viscosity and surface tension (Drosouet et al., 2017) and it necessitates highly refined components. Electrospinning is a type of electrospinning in which the polymeric solution sprays from the nozzle without producing a Taylor cone. Protein electrospinning is possible if the proteins are highly soluble and can form a random coil shape (Bhardwaj & Kundu, 2010). Electrospinning is notoriously challenging for proteins with complicated secondary and tertiary structures. Globular proteins, such as those found in soy, interact too little with one another during the spinning process to entangle (Nieuwland et al., 2014). In biomedical applications, these proteins are electrospun with carriers such as poly (vinyl alcohol) (PVA) or poly (ethylene oxide)

(PEO). Due to the presence of PVA or PEO, no additional solvents are required to dilute the soy protein in an aqueous solution and allow electrospinning (Ramji & Shah, 2014). Unfortunately, certain components are not suitable for use in food. After solubilization in 70% ethanol, zein, the storage protein of maize, was found to be spinnable among the plant-based proteins (Nieuwland et al., 2014). Many spun fibrils, however, are water-soluble and their solubility is reduced by cross-linking with a variety of food-grade reagents, such as phenolic chemicals or the enzyme transglutaminase (Tavassoli-Kafrani et al., 2017). Wheat gluten has been electrospun both alone and in conjunction with polymers (Fabra et al., 2015ab). However, fresh research on replacing spinnable animal-based carriers, such as gelatin, is needed to generate a structurally interesting plant-based meat equivalent and the fibers must also be aligned to make a structurally fascinating food product (Nieuwland et al., 2014). Electrospinning of proteins has been documented for several animal-based proteins such as whey, collagen, egg and gelatin but very sparsely for plant proteins (Ghorani & Tucker, 2015).

1.5.3 Freeze Structuring

An aqueous solution or slurry of proteins is frozen to develop structure in freeze structuring or freeze alignment. Meat, fish and plant protein have all been investigated using directional freezing (Girija et al., 2021). With the freezing temperature, the size of the ice crystal needles can be customized. The frozen product is then dried without melting the ice crystals, resulting in a porous microstructure with sheet-like parallel protein orientation. These sheets are joined together to form a fibrous, cohesive product similar to meat (Girija et al., 2021). Dekkers et al. (2018) in his study explained some of the well-known methods like extrusion, freeze structuring, electrospinning, in vitro culture of animal muscle cells and shear cell technologies for creating the structure of fibrous plant protein materials. Directional freezing has been used to study the structure of meat, fish and plant proteins, as well as to create porous metal and ceramic materials. Proteins must be relatively soluble prior to freezing in order to produce different fiber products and these proteins become insoluble throughout the freezing process (Dekkers et al., 2018). Yuliarti et al. (2021) used a novel freeze structuring technique to test several plant-based composites in order to generate plant-based meat analogues

with unique texture profiles. The analogues physicochemical and sensory features were thoroughly investigated. This analogue's microstructure revealed a fibrous and layered structure. The quantity of cross-linking between protein molecules influenced the analogous textural profiles and structure, which were found to be connected to their viscoelastic capabilities. His research contributed to a better understanding of composite effects in the structure of plant-based meat analogues as well as their physicochemical qualities. The biggest disadvantage is that different freezing conditions must be monitored and optimized at the same time. The innovative freezing process has a positive impact on the production of improved microstructure and quality features (Zhan et al., 2018).

1.5.4 Shear Cell Technology

Shear cell technology is a pristinely incipient approach predicated on the flow-induced structuring principle. Shear cell is a cone-cone device based on a cone-plate rheometer where the upper cone remains motionless and the bottom cone rotates (Grabowska et al., 2016). With the use of an oil bath, both cones can be heated and cooled. In contrast to extrusion, the deformation inside the device during processing is clearly defined and consistent. Proteins are aligned by a mixture of simple shear and heat, resulting in fibrous meat substitutes (Girija et al., 2021). A steamer or a heated bath is used to control the processing temperature. Simple shear flow and heat on the dough result in well-defined fibrous structures that are stable after chilling and do not distort (Manski et al., 2008). Peighambardoust et al. (2007) developed a shear cell device that was effectively used to prepare meat analogues at high temperatures without losing structural conformance, as is common in extrusion. This method was used to treat anisotropic fibers in order to blend a pea protein-wheat gluten and soy protein-wheat gluten mix. Similar to chicken fiber, the resulting fibers have a mechanical strength of 50–100 kPa. Certain factors like processing time, temperature and shear rate can produce changes in shear-induced structuring. This is a low-cost method of manufacturing fibrous meat mimics that are larger in size and have a soft texture (Krintras et al., 2015). More research is needed to scale up this technique for commercial usage.

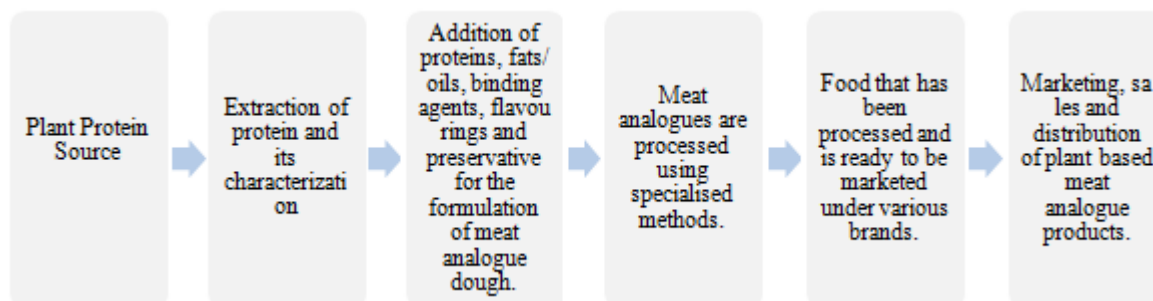


Figure 2: Representation of steps involved in the production of meat analogue

1) Role of meat analogue in vegetarian diet

Due to customers growing health concerns and related environmental challenges, vegetarian foods now occupy a bigger than ever shelf space in today's market (Kumar, 2016). Vegan diets are becoming increasingly popular

among teenagers and young people, particularly among the females. Many vegans base their nutritional decisions on environmental concerns, ethical concerns about animal welfare, the use of antibiotics and growth boosters in the production of animals, the threat of animal-borne diseases

and the health benefits of a plant-based diet (Craig, 2009). A real vegetarian abstains from eating any flesh items, including fish and poultry and instead focuses on fruits and vegetables, grains (ideally whole grains), legumes and nuts. Raw foods are emphasized in several vegetarian diets (Craig, 2010). Processed vegetarian foods (meat analogues, veggie burgers, non-dairy milks and vegetarian entrees) have seen tremendous growth in the United States during the last decade. Vegetarians' nutrient consumption is predicted to be significantly impacted by the widespread availability of fortified vegetarian foods (such as soymilks and meat analogues) (Craig, 2010). Vegan diets tend to be richer in dietary fiber, magnesium, folic acid, vitamins C and E, iron and phytochemicals (Craig, 2009). Vegetarians have a lower risk of cardiovascular disease (CVD), obesity, type 2 diabetes and some malignancies in general (Craig, 2009). However, some studies have revealed that a vegetarian diet can cause negative effects on the body. Hyperhomocysteinemia, protein shortage, anemia, decreased creatinine content in muscles and menstruation disruption are all possible side effects of increased physical activity in women. Some of these changes may impair one's capacity to engage in physically demanding activities (Pilis et al., 2014). Vegans diets exhibit lower blood levels of saturated and monounsaturated fats, cholesterol, fiber, ascorbic acid, folic acid, copper and manganese than meat-eaters. Vegans also had lower amounts of leukocytes, enterocytes, platelets and urea in their blood than meat-eaters, but a higher concentration of albumin. These could point to a vegetarian diet with a decreased protein level. Vegetarians are also in danger of iron, calcium, zinc, vitamin D and B12 deficiencies as well as amino acid shortage (Pilis et al., 2014). It has been found that the inclusion of meat analogues in a vegetarian diet can address this issue. Meat analogues have been shown to have a variety of different proteins as shown in table 1 depending on the protein source used. This can help in the countering of the protein deficiency faced by vegetarians. Proteins derived from legumes, nuts, seeds and whole grains can also be used to augment vegetarian diets. As a result, a properly designed vegetarian diet should give an average of 12.5 percent protein-derived energy (Pilis et al., 2014). As for the concerns regarding other key nutrients which are commonly found to be deficient in a common vegetarian diet, tables 2 and 3 have shown that meat analogues contain iron, dietary fiber, carbohydrates, saturated fats and mineral content which is greater or on par with common meat products. Pawlak et al. (2013) in his article about the study of vitamin B12 stated that meat analogues derived from soy-based products are found to contain high amounts of vitamin B12. A vegan diet appears to be beneficial for enhancing preventive minerals and phytochemicals while reducing dietary variables linked to a variety of chronic illnesses (Craig, 2009). Before looking at consumer views regarding meat analogues, it's important to understand who the meat analogue consumers are on vegetarian, vegan and flexitarian diets (Kołodziejczak et al., 2022).

Davitt et al. (2021) in his investigation looked into differences in spirituality, vegetarianism, environmental sustainability, environmental consciousness and nutrition among eaters and non-eaters of plant-based meat analogues among other things. People who did not consume meat

analogues were more likely to consider themselves religious, according to the findings (Davitt et al., 2021). They were also more inclined to agree that a vegetarian supper isn't a full meal and that vegetarians are "a little different." Those who consumed meat substitutes were more likely to agree that these items were less detrimental to the environment and provided an adequate amount of protein (Davitt et al., 2021).

2) Spoilage and shelf life of meat analogues

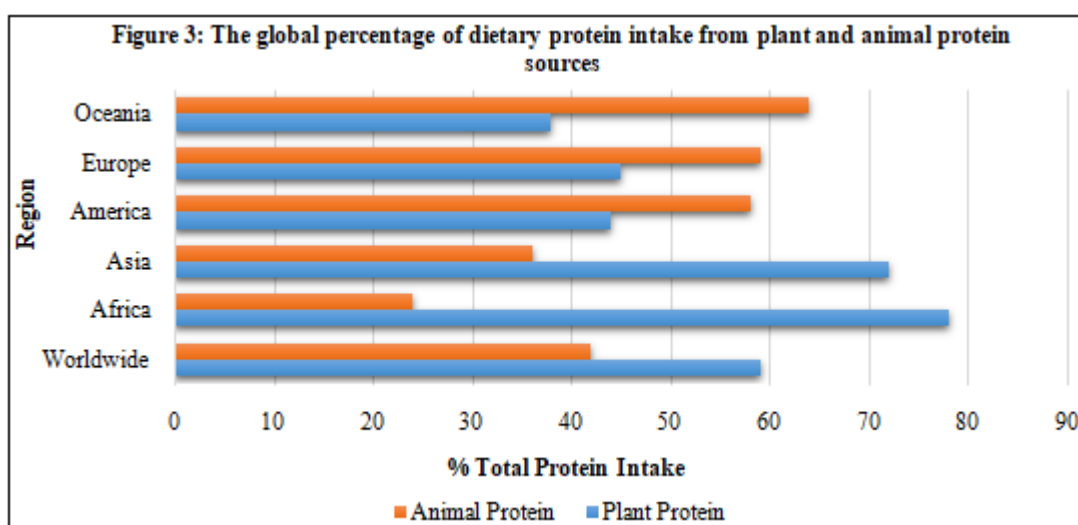
Meat analogues are extremely sensitive to spoiling due to their almost neutral pH and high protein and moisture content (Wild et al., 2014). Microbiological activity is the most important component, followed by microbial enzymes and metabolites. There is a scarcity of information in the scientific literature about the microflora of high-moisture meat analogues made from plant proteins. To detect and characterize important deteriorating and possibly dangerous bacteria, a complete microbiological study of the components and newly created products is required (Wild et al., 2014). Microorganisms cannot grow in powdered plant-based proteins due to their poor water activity. Endospore-forming bacteria on the other hand, maybe able to survive the extrusion process (Schaffner et al. 2005). Pre-mixtures and recipes may contain suitable concentrations of microorganisms depending on the microbial load of the specified raw materials. This includes the existence of potentially harmful and dangerous microbial species. During a normal culinary extrusion process, vegetative microbial cells and bacterial endospores are inactivated to a high degree depending on the process temperature and pressure, as well as the water content. As a result of the microbiological quantification investigation in meat analogue products, it was shown that post extrusion thermal treatments are required to extend the commercial shelf life (Wild et al., 2014). While further sterilization was given to intermediate and final meat substitutes, microbial activity was significantly reduced when held at 6°C for long periods of time (Wild et al., 2014). In conclusion, the expiry date of intermediate goods from the "Like Meat" project that has not been subjected to additional heat and preservative treatments, is comparable to that of consumable meat (Wild et al., 2014). All post-process stages are crucial for product cleanliness and the danger of re-contamination from handling and packing the meat equivalent must be avoided. As a result, alternative post-packaging procedures for final goods, such as sterilization, pasteurization and freezing should be considered (Wild et al., 2014). Thus, it can be concluded that the storage and handling of meat analogues are same as to that of raw meat (He et al., 2020).

3) Market prospects and popularity

Nowadays, there has been a huge demand for meat analogues in the market due to their immense benefits. In the past, consumers found it hard to shift from the meat diet consumption mainly due to their insecurities and doubts about meat analogues providing all the characteristics of meat. Without a question, the popularity of meat analogues is on the rise as more customers pursue protein substitutes and environmentally friendly foods (Ismail et al., 2020). Germany, France, Netherlands, United Kingdom, Italy and Sweden are amongst the top countries in alternative meat protein research and development, with Europe dominating

the global market for meat substitutes. Replicating the meat alternative will break the market in the coming years due to big companies eager to increase their market share as demand for meat alternatives soars (Ismail et al., 2020). According to estimates of meat substitute sales in the United States, UK, Germany, Italy, France, Netherlands, Sweden and Belgium in 2018, plant-based alternatives could rise at a compound annual growth rate (CAGR) of 10% by 2029, totaling to \$1.4 trillion (Ismail et al., 2020). Not only are healthier diets driving the move toward meat alternatives, but also the rising millennial, access to knowledge about food sources, animal welfare concerns and environmental effects. Throughout recent years, the number of vegetarians and consumers who are decreasing their meat intake has increased in Europe. Depending on the sort of customer, environmental, ethical and health considerations are primarily to blame. Other factors include a rise in customer desire for variety and “new” meals. As a result,

one strategy to achieve a significant reduction in meat consumption is to produce meat mimics that compete directly with meat products (Wild et al., 2014). Another option is to launch protein goods that address the desire for additional variety and/or new items. A graphical comparison of the consumption of plant and animal proteins in various regions is shown in Figure 3. The market for meat substitutes is relatively modest. In the Netherlands for example, meat replacements account for just approximately 1% of the entire market for meat and meat products, owing to the fact that current meat analogues do not fulfill customer expectations for sensory quality. In comparison to meat, the bite, flavor and juiciness are all low. Meat analogues must perform better than genuine meat in order to gain a bigger market share. The technique of looking for new goods is particularly well suited to the creation of consumer-oriented items (Wild et al., 2014).



4) Consumer awareness of meat analogue

Consumer awareness has increased as a result of rising food demand and the resulting effect of meat production (Steinfeld et al., 2006). Food security, deforestation, pollution and other environmental and social challenges are additionally becoming major concerns. Consumers are increasingly seeking for sustainable foods and ecologically convivial production techniques, which is emboldening them to go vegetarian or cut down on their weekly meat intake (Dagevos & Voordouw, 2013). Figure 4 shows a representation of the key factors for the acceptance and rejection of plant-based meat analogues among consumers.

Reduced meat consumption reduces the demand for natural resources (water, feed, etc.) and the emissions connected with meat production. As a result, the advantages of converting to a plant-based diet that is nutritionally and sensually equivalent to a meat-based diet appear to be numerous. Several environmental studies on protein-rich foods have been conducted, including plant-based meat analogues, animal-produced proteins and mycoproteins. Many of these studies have ascertained that meat substitutes made from plants have a lesser environmental impact than that of meat (Van Mierlo et al., 2017).

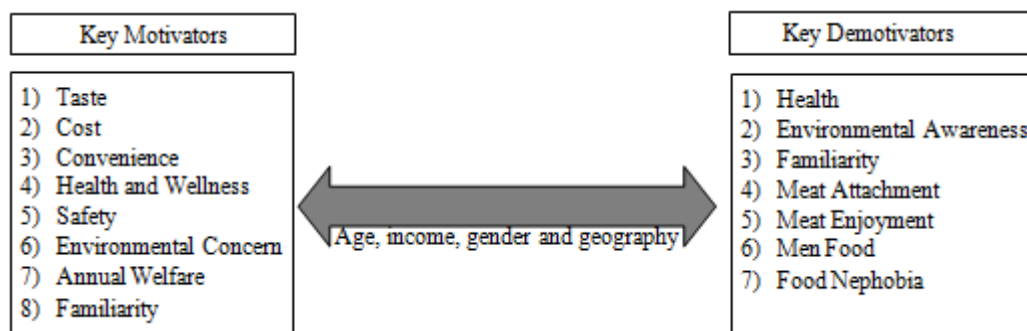


Figure 4: Factors behind consumers' acceptance to purchase plant-based meat analogues

2. Challenges and Future of Meat Analogue

Despite the fact that food technologists and nutritionists face numerous technical challenges in transforming plant proteins into a convenient and suitable meat substitute that appeals to meat-eaters, there have been significant advances in plant protein structuring and formulation. It has been possible to create products that look like meat or have the overall appearance of certain meat pieces (Fonmboh et al., 2021). Plant-based meat analogues are thought to be an excellent way to improve human health, protect natural resources and maintain animal welfare. Many culinary components, in addition to their nutritional value, have the capacity to prevent and cure diseases, therefore they come into the category of medical food homology (MFH) (Sun et al., 2020). Other than thermal and mechanical treatments like the extrusion process, the “bottom-up” method could be applied for the production of meat analogues. This way, we can now enjoy tasty, nutritious meals without causing harm to the environment thanks to the growing number of choices for meat analogue products (Sun et al., 2020). Because of its intrinsic qualities as a very cheap source of protein, meat analogue has a promising marketing future. It is ideal for health-conscious non-vegetarians, lactose-intolerant people and people observing religious guidelines or to fix ethical qualities and nutritional problems for vegetarians (Kumar et al., 2016). As a result, meat analogues have a much greater chance of success than other products, since some consumers want a food that is organoleptically appealing and healthy but is free of meat.

In many nations, current meat consumption levels not only exceed dietary protein requisites but are also unsustainable (van der Weele et al., 2019). In terms of environmental, health and ethical concerns, the health-promoting dietary components will be an excellent alternative for meat consumption. The use of protein-rich meat analogues helps to combat climate change and diverts attention away from the unsustainable meat business, which produces a lot of waste, pollutes the environment and makes animals vulnerable to illnesses. Many nutritional quality, shelf-life, taste and flavor issues have already been addressed thanks to appropriate advancements in plant-based meat food items and marketing techniques (Sha & Xiong, 2020). Hence, the concise and precise study of the development of meat analogue is a necessary aspect which is what we are going to look through. The development of meat analogue doesn't just stick inside a certain shell but it spreads across a vast pool of different aspects.

3. Conclusion

The concept of meat analogue and its prevalence are increasing and can be attributed to the consumer's increasing concern for health and interest towards nutritional food diets. In terms of nutrition, health and benefits, meat analogue can be considered as a suitable replacement for meat and meat products. As many consumers regard meat products to be an essential component of their diet, the argument on meat and health may also be in support of meat intake. The most significant factors in deciding whether or not to consume meat are its taste and nutritional value. Developing meat substitutes that are nutritionally and

physically similar to meat can improve people's willingness to forego meat intake. However, extra non-protein components are employed to imitate meat in other sensory qualities such as color, flavor and texture. Although there are still some questions about protein requirements, research from adult vegetarians suggests that traditional vegetarian diets provide more than enough protein and amino acids. There may be a small risk of insufficient intake in a small percentage of vegans and more data is needed to analyze the real dietary pattern of those who report dietary intake that corresponds to a low intake of protein and calories. Product development is hampered by the wide range of component functionality requirements across different types of plant-based meat substitutes. Meat analogues will have to address against these claims and the reasons for its commercialization and popularization in the market.

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