The Flavor of Plant-Based Meat Analogues

Xuejie Li¹ and Jian Li¹,²,³

ABSTRACT

Plant-based meat analogues are important protein sources due to their high nutritional value and low resource consumption. The flavor of meat analogues is a key factor in consumer acceptance. The process of imparting meat-like flavor in plant-based meat analogues is very complex, requiring analysis and identification of the flavor compounds produced by the Maillard reaction, lipid oxidation, and thiamine degradation of animal meat products. Some vegetarian ingredients, such as hydrolyzed vegetable protein, yeast extracts, natural spices, and certain vegetable oils, have been applied in meat analogues to simulate meat-like flavors.

Meat is a highly nutritious, health-promoting food and an excellent source of minerals, heme iron, and essential amino acids (1). Additionally, meat proteins contribute greatly to the food industry by imparting specific functionalities, such as gelation, emulsification, and water-holding properties (2). The appearance, texture, and flavor of meat products depend on their protein properties, which are difficult to substitute for using alternative food ingredients (3).

In 2017 the World Health Organization predicted global per capita meat consumption will increase to 45.3 kg/year by 2030, and the meat industry will need to increase production by approximately 50–72% to meet the calorie and nutrient requirements associated with population growth (4). However, animal meat products are ecologically burdensome and resource-intensive and are foods that raise ethical concerns (5). Moreover, zoonosis, strong demand for “healthy” foods, economic constraints (e.g., rising meat prices), and the increasing popularity of vegetarianism have stimulated the development of meat analogues based on plant proteins (6). Plant-based meat analogues simulate the appearance, texture, flavor, and functionalities of traditional meat products but differ in composition (2).

Texture is one of the most important aspects of meat products. However, flavor is also an increasingly important aspect for consumers (7). Currently, most plant-based meat analogues are formulated with wheat gluten, soy, or pea proteins because of their lower costs and excellent properties, such as oil absorbency, gelation, emulsification, and water-holding (8). These plant proteins impart a fibrous texture that is similar to meat through high moisture extrusion. To develop meat-like flavor, flavor compounds from vegetarian ingredients are extracted and analyzed using several methods, including solid-phase microextraction, solvent-assisted flavor evaporation, and gas chromatography–olfactometry–mass spectrometry (9). Vegetarian ingredients with meat-like flavor are added to the texturized plant protein. Many researchers have performed studies on the texture properties of plant-based meat analogues and have developed several technologies, such as a shear-induced technique, spinning, 3D printing, and extrusion, to improve palatability (10). However, only a few studies have reported on the flavor properties of plant-based meat analogues. In this review, we focus on ingredient sources and the pathways to formation of flavor compounds in plant-based meat analogues.

Mechanism of Meat Flavor Formation

Like texture and aroma, flavor is a critical sensory attribute of food products (11). To impart meaty flavor in plant-based analogues, we must understand the mechanisms of the formation of flavor compounds in animal meat. For example, there is a great difference between the flavors of raw versus cooked meats. Raw meat has no aroma and only the flavors of blood, metal, and salt, while cooked or roasted meat products have an aroma (12) and the flavor compounds become more complex as processing temperatures increase (13).

Meat is composed of water, carbohydrates, lipids, proteins, vitamins, and minerals. Among these components, carbohydrates, proteins, and fat play a major role in flavor development, because they contain many vital flavor precursors (7). When heated, various volatile flavor compounds (e.g., alkenes, alcohol, aldehydes, ketones, ethers, esters, carboxylic acids, sulfur-containing compounds, etc.) are produced by the reactions of flavor precursors in the meat (14,15). The flavor of the meat arises from inorganic salts, short peptides, free amino acids, and nucleic acid metabolites, such as inosine and ribose, and aroma is derived from volatile flavor compounds, such as unsaturated aldehyde ketone, heterocyclic compounds, and sulfur-containing compounds, which generally are produced by heating (16).

The flavor compounds in cooked meat are produced by extremely complex decomposition, oxidation, reduction, and other chemical reactions (Table I). Primary reactions include 1) the Maillard reaction between amino acids and reducing sugars; 2) the oxidation of fatty acids; and 3) the thermal degradation of thiamine (16). Presently, more than 1,000 volatile compounds have been identified from these reactions (13).

Table I. Flavor-forming precursors, thermal reactions, and key flavor compounds in meat products

<table>
<thead>
<tr>
<th>Flavor Precursors</th>
<th>Thermal Reaction</th>
<th>Key Flavor Compounds</th>
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</thead>
<tbody>
<tr>
<td>Reducing sugars, free amino acids, peptides</td>
<td>Maillard reaction</td>
<td>Pyrazine, heterocyclic compounds, sulphydryl compounds</td>
</tr>
<tr>
<td>Lipids, fatty acids</td>
<td>Oxidation</td>
<td>Aldehydes, furans, unsaturated ketones, aliphatic hydrocarbons</td>
</tr>
<tr>
<td>Thiamine</td>
<td>Degradation</td>
<td>Thiols, sulfides, disulfides</td>
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1 Beijing Engineering and Technology Research Center of Food Additives, Beijing Technology and Business University, Beijing 100048, China.
2 Beijing Advanced Innovation Center for Food Nutrition and Human Health, Beijing Technology and Business University, Beijing 100048, China.
3 Corresponding author. Business address: No. 33 Fucheng Road, Beijing, China. Fax: +86-10-68984032; E-mail: lijian@btbu.edu.cn

Adapted from Issa Khan et al. (18).
Maillard Reaction. The Maillard reaction is an important nonenzymatic browning reaction that takes place during food processing. It includes complex reactions, such as polymerization and condensation, at high temperatures between compounds containing free amino groups and carbonyl compounds (also termed carbonyl ammonia reaction) (19). The specific Maillard reaction process is shown in Figure 1.

According to Hodge’s model, the Maillard reaction contains three reaction stages, and each stage is divided into several reactions (20). In the initial stage, Amadori rearrangement occurs between aldoses and amino acids, forming Amadori rearrangement products (1-amino-1-deoxy-2-ketose), which can initiate further reactions (21). In the intermediate stage, Amadori rearrangement products undergo Strecker degradation. Strecker degradation produces 2-aminocarbonyl compounds and Strecker aldehydes, which are important intermediates during the formation of meaty flavor (22). In the final stage, the intermediates of the previous two stages, such as ketones and aldehydes, aggregate with amino compounds to form complex insoluble brown polymers, called melanoidins (23). Moreover, the aminoketone is cyclized, which generates pyrazine compounds.

Many precursors of the Maillard reaction, such as reducing sugars (xylose, ribose, glucose), amino acids (methionine, cysteine, threonine, glycine, alanine, serine), flavor nucleotides (5′-IMP and 5′-GMP), and thiamine, can mimic meat aromas in plant-based meat analogues (2). The reducing sugars in meat arise mainly from the breakdown of glycogen and partly from energy metabolism (e.g., ribose—a product of ATP decomposition) (19). Additionally, many studies have shown that phosphorylated monosaccharides are more prone to undergoing Maillard reaction than are nonphosphorylated monosaccharides (24). Among the amino acids involved in the Maillard reaction, cysteine is considered the most critical amino acid for generating meat-like flavor (19). Consequently, the system of cysteine and ribose is important in the generation of meaty flavor (25) in plant-based meat analogues. More than 180 compounds, including thiols, polysulfides, and thiophenes, have been identified in these reaction systems (7), and sulfur-containing compounds are crucial to the formation of meaty flavor. Because xylose is a less expensive isomer of ribose, it is often used in manufacturing plant-based meat analogues. In the cysteine and xylose system, relatively stable cyclic 2-threitylthiazolidine-4-carboxylic acids are formed (21), which are also related to meat-like flavor compounds.

Oxidation of Lipids. Generally, the precursors of meaty flavor compounds can be classified as lipids and water-soluble compounds in the muscle tissues. Water-soluble compounds in muscle tissue are mainly involved in the Maillard reaction, whereas lipids undergo an oxidation reaction. The characteristic flavors of different kinds of meat (e.g., beef, pork, poultry, fish) depend largely on the volatiles produced by the oxidation of lipids. These volatile flavor compounds possess a higher odor threshold (flavor compounds with higher thresholds are more difficult to smell) than compounds generated in the Maillard reaction.
reaction, but they can significantly influence the total flavor profile because of their abundant content (26).

Some studies have shown that in addition to the conditions encountered in intense barbecue heating, the amounts of volatile compounds produced by the oxidation of lipids account for the majority of the total flavor compounds (27). In addition, certain fatty acids produced by the oxidation of lipids can react with the intermediate compounds of the Maillard reaction, forming flavor compounds with lower odor thresholds (26). Furthermore, lipids can act as solvents to increase several aroma compounds during cooking and processing of meat (28). Phospholipids and triglycerides are the major components of animal lipids, and phospholipids are the main contributors to meaty flavor due to their higher degree of unsaturation (28). Phospholipids contain many unsaturated fatty acids (UFAs; mainly polyunsaturated fatty acids [PUFAs]), which break down into flavor compounds through the primary and secondary stages of oxidation (29) (Fig. 2). In the primary phase, UFAs undergo the initiation, transport, and termination phases of the chain to eventually form hydroperoxides (30), which are odorless substances. In the secondary phase, these hydroperoxides are thermally oxidized at temperatures higher than 150°C, producing hundreds of volatile compounds, such as aldehydes with 6–10 carbon atoms, furans, unsaturated ketones, aliphatic hydrocarbons, etc., that contribute to a desirable meaty flavor.

In contrast, nonthermal lipid oxidation reduces meat quality, causing rancidity, off-odors, and color changes in meat (31). The process produces different aldehydes (heptanal, hexanal, glutaraldehyde, etc.) that produce disagreeable odors even at very low concentrations (32).

**Thermal Degradation of Thiamine.** Thiamine, a sulfur- and nitrogen-containing bicyclic compound, is thermally degraded, producing a multitude of sulfur and nitrogen compounds, such as thiols, sulfides, and disulfides (34). Hydrogen sulfide is not a flavor compound, but it can react with ketones to form volatile compounds with a strong meaty flavor. For example, 2-methyl-4,5-dihydroxy-3/4-thiophenethyl and 2-methyl-2,3-dihydroxy-3/4-thiophenethyl contain the distinct flavors associated with roasted or boiled beef. 2-Methyl-3-furanthiol (MTF), a critical precursor of various thioether compounds related to meaty flavor, is generated by the Maillard reaction between ribose and cysteine or the Strecker reactions of sulfur-containing amino acids (35). However, some researchers found that amounts of MTF increased four- to fivefold when they added small amounts of thiamine to the Maillard reaction system between ribose and cysteine (36). This indicates the thiamine is degraded to produce 2-methyl-3-furanthiol more efficiently even in low concentrations than in the Maillard reaction.

**Challenges of Producing Meat-like Flavor in Meat Analogues**

Plant-based meat analogues consist mainly of soy protein and wheat gluten, which do not contain the key intermediate compounds needed to produce a meat-like flavor. Therefore, various meat-like flavorings are added to meat analogues to mimic the flavors and aromas of meat. However, some synthetic flavors added to meat analogues reduce the quality of the product and generate potentially harmful components. In addition, the off-flavors of certain plant proteins negatively influence the formation of meat-like flavor in meat analogues.

**Disadvantages of Synthetic Flavorings.** To simulate the flavor of animal meats, many synthetic flavors are added to conventional plant-based meat analogues. In some foods they can impart vivid original flavors, strengthen aroma, and mask off-flavors (37). Generally, flavoring agents can be divided into chemical synthetic and natural agents. Synthetic flavoring agents are produced using chemical methods.

Recently, some synthetic flavoring agents have been used as food additives in conventional meat analogues. However, the addition of synthetic flavoring agents can create some challenges.

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**Fig. 2.** Oxidation of fatty acids in processed meat. Adapted from Ji et al. (33).
There are different flavors associated with different types of meat products, which are mainly related to lipids in specific animals (e.g., beef, pork, poultry, fish). The common synthetic flavoring agents lack the characteristic flavors of specific types of meat. Also, a synthetic flavoring does not last long and can be easily destroyed during cooking. Hence, the flavor changes experienced between raw and cooked meats cannot be replicated by synthetic flavorings. Moreover, synthetic flavorings may have adverse effects on human health, accumulating in the human body over a long period of time and leading to the development of toxic compounds, such as carcinogens, mutagens, and teratogens (38).

**Off-flavors in Soy-Based Meat Analogues.** Soy protein isolate (SPI) is the main raw material used in meat analogues and has the advantages of high nutritional quality, broad supply source, and low cost. However, SPI produces an undesirable off-flavor, which is a major drawback to its utilization in plant-based meat analogues (39). This off-flavor is described as beany, grassy, chalky, bitter, and astringent (40) and is caused by two factors: the astringent and bitter flavors are produced by isoflavones and saponins in soy protein, and the grassy and beany flavors are formed due to peroxidation of UFAs, such as linoleic and linolenic acids, by lipoxygenase (LOX) (41).

Raw soybeans contain about 20–23% lipids, which mainly include monounsaturated fatty acids, such as oleic acid (18:1), and PUFAs, such as linoleic (18:2) and linolenic (18:3) acids (39). LOX has high enzyme activity, and it can react with lipid substrates when the soybean cell wall is broken. The reaction can be promoted by suitable metal ion catalysts, such as Fe$^{2+}$ and Cu$^{2+}$. The main volatile off-flavor compounds produced by lipid oxidation include short-chain fatty acids, aldehydes, and ketones. These nonvolatile compounds include long-chain alcohols and esters (40). To address these challenges, the flavor of a legume protein should be analyzed to determine how undesirable flavors can be reduced. Recently, various methods, including acid treatment, solvent treatment, enzyme decomposition, and genetic engineering, have been applied to remove the off-flavor in soy protein.

**New Approaches to Creating Meat-like Flavor**

Natural flavoring agents are generally more acceptable than synthetic ones. They are commonly extracted from natural sources, such as aromatic plants or raw animal materials, using physical methods and are safer for human consumption. However, most natural meat-like flavorings contain meat protein hydrolysates and animal lipids, which is not acceptable to vegetarians. Consequently, plant-based ingredients, such as hydrolyzed vegetable protein (HVP), yeast extract (YE), and natural spices, are widely used in the formulation of plant-based meat analogues.

**HVP.** HVP, a nutritional food additive, has been used for more than a hundred years to impart meat-like flavor in foods (42). More recently, HVP has been utilized in the production of meat analogues. It can be used not only as a flavorant, creating its own flavor, but also as a flavor enhancer that promotes the natural flavor of a food product (43).

HVP is prepared from a variety of plant proteins, including soybean, corn, wheat, peanut, etc., which are broken down into amino acids and short peptides by hydrochloric acid or enzymatic hydrolysis (44). However, the application of hydrochloric acid can form potentially carcinogenic compounds, such as 3-chloropropane-1,2-diol (42). Enzymatically hydrolyzed vegetable protein is produced using proteolytic enzymes under milder pH and temperature conditions (45). HVP itself contains many volatile aroma components, such as pyrazines, pyridines, pyrrole, organic acids, furans and furanones, sulfur-containing compounds, alcohols, aldehydes, ketones, esters, and phenols (46). When coheated with sulfur compounds, reducing sugars, and yeast autolysis, they produce a strong meat-like flavor.

Zhao et al. (47) hydrolyzed rapeseed protein with trypsin and a neutral protease (a special type of protease extracted from *Bacillus subtilis*) and then added some substrates of the Maillard reaction to produce meat-like flavor. They found that the optimal reaction conditions were a pH of 6.0, reaction temperature of 120°C, reaction time of 60 min, 6.0% xylose, 1.2% cysteine hydrochloride, 0.8% methionine, and 0.1% thiamine. As an important intermediate of the Maillard reaction, mercaptacetaldehyde is produced by Strecker degradation of cysteine hydrochloride, which breaks down further, producing acetaldehyde and hydrogen sulfide and imparting a meaty aroma. However, when the concentration of cysteine hydrochloride increases, the excess hydrogen sulfide produces an undesirable flavor. Methionine can generate disulfide compounds, which is necessary for the Maillard reaction.

**YE.** YE, a natural seasoning, is prepared from food-grade yeast, which has high protein and nucleic acid contents (e.g., brewer's yeast and baker's yeast). Yeast itself contains some volatile flavor compounds, such as thiophene and pyrazine, which can produce a roasted meat aroma that is sweet and fragrant (48). YE utilized as a flavor enhancement is enriched with nonvolatile flavor precursors, including reducing sugars, amino acids, peptides, nucleotides, lipids, and thiamine, which contribute to meaty flavor only upon heating (49). It is the combination of these volatile and nonvolatile compounds that forms the meat-like flavor in YE (50).

Some researchers have identified aromatic active compounds in YE. Lu et al. (51) prepared meat-like flavorants from brewer's YE without the addition of animal lipids and meat protein hydrolysate. Through extraction and sensory identification, Lin et al. (48) identified some key aroma-active compounds in YE, including 2,3-butanediol, acetic acid ethyl ester, 3-methylbutanal, and 2-acetyl-1-pyrroline, which impart a roasted meat flavor. These compounds originate from the Maillard reaction, lipid oxidation, and thiamine degradation in YE, which is similar to the formation pathway of flavor compounds in meat. In addition to the meat-related flavor, YE is also associated with umami and kokumi flavor constituents, such as aspartic acid, glutamic acid, and 5'-nucleotides (52).

**Natural Spices.** Natural spices are natural plant ingredients with fragrant, spicy, hemp, bitter, and sweet flavors. Each natural spice has a unique flavor and aroma. For example, Sichuan pepper exhibits a pungent flavor (53), and *Amomum villosum* imparts a delicate aroma. In addition to imparting unique flavors in meat products, natural spices can enhance natural flavors, reduce undesirable flavors, highlight the typical flavor of a food, and help food flavors coordinate. Natural spices commonly used in meat products are shown in Figure 3. Some of them can be added to plant-based meat analogues in the form of powders or extracts to improve flavor and enhance sensory properties. Moreover, some of these spices contain antioxidants that can protect the lipids and oils in meat analogues against oxidative degradation (54).

Garlic oil has been used in the production of meat analogues in recent years. In 1948, Stoll and Seebeck used ethanol as a solvent to extract the precursor of characteristic garlic flavor com-
pounds, “alliin” (55), which can be degraded into alkyl sulfides such as diallyl trisulfide, diallyl sulfide, and diallyl disulfide (56), which are associated with the formation of meat-like flavor compounds.

Onion powder is also utilized in meat analogues. Thiosulfonate is the most abundant flavor precursor in onion and is derived from the degradation of S-alk(en)yl-L-cysteine-S-oxide (56). Similar to the flavor precursor in garlic, thiosulfonate is a very unstable compound, and it can easily rearrange to produce different sulfides that can form meat-like flavor compounds.

Vegetable Oils. Lipid oxidation produces meaty flavor, which contributes to tenderness, juiciness, and flavor release in a meat product (57). The addition of fat or oil to plant-based meat analogues is considered necessary because it can oxidize itself to produce flavor compounds and also can prevent the loss of other flavor components during processing (58). Some vegetable oils, used in meat analogues include coconut, pressed canola, and sunflower, which are used to simulate the role of animal fats in flavor formation. Analysis and identification of flavor compounds have shown that these vegetable oils contain fatty acids similar to animal lipids. These oils are mixed in proportion and oxidized directionally at a specific temperature to simulate the flavor of animal lipids. For example, currently available meat analogues (e.g., Impossible Meat) contain only 12–15% fats because the addition of high amounts of fat can affect the formation of fibrous structures.
Conclusions

Plant-based meat analogues should be sustainable products with functional characteristics and sensory properties (i.e., appearance, texture, aroma, and flavor) similar to animal meat. At present, flavor is one of the most important aspects of meat analogues and the key to consumer acceptance.

In this review, we discussed strategies for imparting meaty flavor to plant-based meat analogues using plant-derived ingredients (Fig. 4). The flavor of meat analogues is developed in three parts. In the first step, nonmeat protein is enzymatically hydrolyzed to produce the precursors of typical meat flavors. In the second step, vegetable oil is oxidized directionally to simulate the flavor of animal lipids. In the third step, natural spices are added to reduce off-flavors and promote flavor release.

Compared with meat products, plant-based meat analogues are more nutritious and healthier. They can be used to effectively alleviate the shortage of meat resources. Functionality, sensory properties, and costs influence the buying behavior of customers. With regard to sensory properties, researchers have identified the flavor compounds in animal meat through molecular sensory science and technology and are seeking more suitable substitutes from plant ingredients. Although certain technologies for the production of meat analogues still need to be developed, the marketing prospects for plant-based meat analogues are very high.

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