

MEATS

Meats*Fresh Meats*

Of all aromas the generic aroma of cooked meat most stimulates the appetite of a large majority of mankind. The reconstruction of meat flavor for flavoring other food products represents an extremely important goal for the flavor industry. Until recently, however, thoroughly functional flavor imitations have not been formulated. In the continuing investigation of meat flavors, probably more emphasis has been placed on odor (resulting from the most important volatile components) rather than on flavor. Analytical studies of the natural flavor of precooked meat have uncovered similarities in the flavors of red meats from various animals—beef, lamb, pork, etc. The differences between these and poultry flavor are considerable.

Red Meats

In beef, lamb, and pork the flavor similarity is only in the "background note." The different flavor sensations experienced when tasting the various red meats are familiar to everyone. All red meats, independent of condiments and seasoning, exhibit a characteristic aroma and flavor that may be classified (tentatively) in relation to the animal source. Several other factors, such as age, type of feed, breeding, etc., strongly affect the resulting aroma of the various meats.

The flavor of cooked red meat (not mentioning the specific characteristics for each type of meat not yet investigated) develops from amino acids, reducing sugars, and fat precursors. Three fundamental reactions involving the above constituents may occur during cooking:

1. Strecker's degradation (deamination and decarboxylation) of β -amino acids, forming the following carbonyl compounds, acids, etc.:

Acetaldehyde	Lactic acid
Acetic acid	Methylamine
Ammonia	3-Methylbutanal
Dimethyl sulfide	Methyl mercaptan
Ethyl mercaptan	2-Methylpropanal
Formaldehyde	2-Methylpropionic acid
Formic acid	Propanal
Hydrogen sulfide	Propionic acid

2. Maillard's reaction, which leads to a darkening of the meat caused by the reaction between amino acids and reducing sugars, with the observed formation of the following alcohols and mono- and dicarbonyl compounds:

Acetone
2-Butanone
Diacetyl
Ethyl alcohol
Methyl alcohol

3. Oxidation of free fatty acids and triglycerides. Fats (which act as a sink* for aromatic substances liberated by heat) yield the following carbonyl compounds:

Acetaldehyde	2-Hepten-1-al
Acetone	Hexanal
Aldehydes, di-unsaturated	2,4-Nonadien-1-al
Aldehydes, mono-unsaturated	Nonanal
Aldehydes, saturated (mainly hexanal)	Octanal
2,4-Decadien-1-al	Propanal
	Undecenal (?)

* It is probable that fats absorb the characteristic aromas exhibited by the above types of meat.

2-Decen-1-al

2-Undecen-1-al

2,4-Heptadien-1-al

The presence of 5'-nucleotides in meat flavor has also been reported.²⁵

Poultry

From the large number of studies of the identification of the constituents of chicken flavor (which consists of approximately 200 components, of which only a few have been identified), one might conclude that the formation mechanism of chicken flavor has not yet been clarified. While the oxidation of lipids is extremely probable, the degradation of amino acids (Strecker's reaction), although highly probable, has not been clearly demonstrated. The latter should occur concomitantly with more complex mechanisms, however.

Chicken flavor consists of a complex chicken-meaty flavor. Characteristic components of the meaty flavor probably consist of sulfur-containing compounds. Components responsible for the chicken flavor are probably the carbonyl compounds that follow:

Acetaldehyde	2-Hepten-1-al
Acetoin	Hexanal
Acetone	2-Hexen-1-al
Butanal	Nonanal
2-Butanone	Octanal
2,4-Decadien-1-al	Pentanal
2-Decen-1-al	2-Pentanal
Diacetyl	Propanal
2,4-Heptadien-1-al	2-Undecen-1-al

The presence of hydrogen sulfide and alkyl mercaptans up to C₆ has been confirmed, while the presence of symmetric and asymmetric alkyl sulfides and disulfides is extremely probable. Esters, hydrocarbons, benzene, and furan derivatives, resulting from not yet clarified mechanisms, have also been reported as probably present.

Preservation of Meats

The preservation of meat has been practiced for centuries as a means of stocking foodstuffs. With the improvement of preservation techniques, the practice has been slowly turned over to the food industry. In the past drying, salting, and smoking were the most common and simple methods used to preserve meat. Air drying was the most commonly used method by primitive people. This method is still practiced today for the preparation of *tasajo* and *pemmican*, dried and salted red meats preserved under corn flour or molten fat.

The preservation of meat is strictly related to the use of flavor ingredients that not only exhibit a flavor action of their own but also enhance the flavoring due to salting. Even in the most simple case of smoking dried or pre-salted meat, not only is the type of wood (birch, maple, olive, etc.) that is burned important to flavoring but also the leaves, berries, etc. of aromatic plants (bay leaf, juniper, *origanum*, etc.) that are burned. Flavor ingredients may also be added during drying; they are added sometimes to the pickling solution prior to drying. The preservation techniques that contribute most toward flavoring are salting and sterilization.

Salt-cured Meats

Salting (sometimes complementary to the drying and smoking operations) is carried out by pickling in a salt solution or by covering the meat with salt. It is used to preserve whole, chopped, or ground meat, either raw or precooked. From a technical flavoring viewpoint, salting is the most interesting operation in the preservation of meat. The addition of salt (1) decreases the free moisture content that is absorbed on the salt as water of hydration, (2) causes continuing dehydration of the meat during slow drying, and (3) improves the organoleptic characteristics during ripening (also because of the addition of about 1% potassium nitrate to salt).

Salt-cured meats may be classified in two broad categories—salted whole meats and cured meats. Salted meats include *prosciutto*, cooked ham, and bacon. Cured meats consist of raw or cooked ground meat, such as salami, sausages, etc. A few salted whole and cured meats, such as *prosciutto*, sausage, etc., also may be smoked.

Each product requires a specific flavoring usually imparted either by maceration in flavored wine, by direct addition of flavor ingredients to ground meat, or by both of these together with the addition of salt. Flavor ingredients are usually spices or ground herbs, oleoresins, and essential oils plated on salt or other similar carriers. Pepper, cinnamon, clove buds, nutmeg, cardamom, coriander, capsicum, and ginger are frequently employed together with garlic, sage, thyme, bay, caraway, and *origanum*.

Smoking may be replaced by a suitable flavoring that, wherever permitted, is effected with cade oil, to which birch, laurel, or pepper oil dispersed in the ground paste or in the maceration solution is sometimes added.

Canned Meats

Canned meats are usually sterilized and require the addition of flavor ingredients. Sterilization is conducted by thermal treatment (120 °C) in autoclaves on shredded, precooked, or raw meat (red and also mixed). Precooked meat is best flavored using a mixture of celery, carrot, laurel, nutmeg, pepper, and garlic tailored to yield a neither too sharp nor too strong final flavor. Flavor ingredients are usually added during preliminary cooking.

MEAT FLAVOR

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INTRODUCTION

The flavor of meat is attributed to a complex mixture of compounds produced by heating a heterogeneous system containing nonodorous precursors, and it is comprised of:

1. Volatile compounds with odor properties.
2. Nonvolatile compounds with taste and tactile properties.
3. Potentiators and synergists.

The chemistry of the compounds responsible for the flavor of cooked meat, and how these flavor compounds are produced during the process of cooking, have been the subject of considerable research in the last two decades. Two obvious facts about cooked meat flavor are

1. The nature of the flavor depends upon the way in which meat is cooked.
2. Each meat has its own characteristic flavor.

Since all types of meat consist of broadly similar components — such as proteins and other amino compounds, lipids, carbohydrates, and mineral salts — it is apparent that there must be a significant difference either in the proportion in which these compounds are present or in the way in which they interact. The differences in flavor due to cooking are probably a direct function of temperature and degree of moisture in the meat.¹ Because it is necessary to heat meat to produce the desired flavor, it follows that meat must contain substances which interact or degrade during cooking to produce compounds responsible for the characteristic flavor of meat. These substances are termed flavor precursors. A general discussion on the importance of flavor precursors and enzymes in the development of various food flavors was given in an earlier review.² This review deals with the nature of volatile and nonvolatile compounds, flavor precursors, and enzymes that may, directly or indirectly, influence the flavor of cooked meat and ripened sausage.

NATURE OF MEAT FLAVOR

Volatile Compounds

The list of volatile compounds isolated from cooked meat is quite extensive and their contribution to the 'typical' flavor of meat uncertain. Tables 1 and 2 list most of the volatile compounds identified so far from beef and pork products. Most of these volatiles are produced during cooking from nonvolatile precursors. Unlike fruit, dairy, and fermented food flavors, enzymes have little significance in the development of cooked meat aroma from precursors.

The nature of volatile compounds responsible for the characteristic aroma of meat has not been resolved. However, significant advances have been made in the last 15 years. Hornstein et al.^{6,7} in 1960 reported the isolation of two major fractions of volatile compounds from a lean beef slurry:

1. Highly volatile fraction containing ammonia, traces of methylamine, hydrogen sulfide, methyl mercaptan, formaldehyde, acetaldehyde, acetone, and other unidentified highly volatile compounds. A rather disagreeable aroma was noticed in this fraction.

TABLE 1
Volatile Compounds Isolated from Heat-treated Beef

Compound	Meat sources and treatment*	References
Acids		
Formic	9	15
Acetic	9,13	15,24
Propionic	9	15
Butanoic	2,6,13	10,16,24
Hexanoic	7	3
2-Methylpropionic	1,9	4,5,15
Aldehydes (aliphatic)		
Formaldehyde	3,6	6,7,10
Acetaldehyde	1,3,4,6,7	3,4,5,6,7,8,10,11
Propanal	1,3,6,7	3,4,5,6,7,10,11
Pentanal	7,8,12,13	3,12,13,14,24
Hexanal	3,7,8,12,13	3,6,7,12,13,14,24
Heptanal	7,8,12,13	3,12,13,14,24
Octanal	7,8,12,13	3,12,13,14,24
Nonanal	3,7,8,12,13	3,6,7,12,13,14,24
Decanal	12,13	24
Undecanal	8,12,13	12,13,14,24
Dodecanal	12,13	24
Tridecanal	13	24
Hexadecanal	7	3
Methional	8,14	12,13,14,25
2-Methylpropanal	1,6	4,5,10
2-Methylbutanal	12,13	24
3-Methylbutanal	1,4,7,8,12,13	3,4,5,12,13,14,24
2-Hexenal	13	24
2-Heptenal	12,13	24
2-Octenal	7,12,13	3,24
2-Nonenal	12,13	24
2-Decenal	12,13	24
2-Undecenal	12,13	24
2-Dodecenal	13	24
2-Tridecenal	13	24
6-Methylhepten-1-al	7	3
2,4-Octadienal	12,13	24
2,4-Nonadienal	13	24
2,4-Decadienal	7,12,13	11,24
2,4-Undecadienal	13	24
2,4-Dodecadienal	12,13	24
Esters		
Methyl formate	2	6
Ethyl acetate	7	3
Acetol acetate	14	25
Ethers		
Pentyl ether	7	3
Pyrroles		
Acetylpyrrole	13	24
1-Methyl-2-acetylpyrrole	14	25

TABLE 1 (continued)

Volatile Compounds Isolated from Heat-treated Beef

Compound	Meat sources and treatment*	References
Alcohols		
Methanol	1	4,5
Ethanol	1,7,8,9	3,4,5,12,13,14,15
Propanol	7,13	3,24
2-Propanol	8	12,13,14
2-Methylpropanol	7	3
Butanol	6,7,8,12,13	3,10,12,13,14,24
3-Methylbutanol	8	12,13,14
Pentanol	7,8,12,13	3,12,13,14,24
Hexanol	8,12	12,13,14,24
Heptanol	8,12,13	12,13,14,24
2-Heptanol	14	25
3-Heptanol	12,13	24
4-Heptanol	13	24
Octanol	7,8,12,13	3,12,13,14,24
3-Octanol	13,14	24,25
4-Octanol	13,14	24,25
Nonanol	14	25
Decanol	14	25
Undecanol	14	25
2-Butoxyethanol	12	24
2-Hexenol	7	3
1-Pentene-3-ol	7	3
1-Octene-3-ol	7	3
2-Octenol	12	24
2,3-Butanediol	12,13	24
Ketones		
Acetone	2,3,4	6,7,8,16
Propanone	1,3,9	4,5,6,7,15
2-Butanone	1,6,8	4,5,10,12,13,14
Acetoin	7,8	3,12,13,14
Diacetyl	5,7,9	3,9,15
2-Heptanone	12,13	24
3-Heptanone	13	24
2-Octanone	12,13	24
3-Octanone	12,13	24
4-Octanone	7	3
2-Nonanone	12,13	24
3-Nonanone	7	3
2-Decanone	12,13	24
2-Undecanone	12,13	24
2-Dodecanone	13	24
3-Dodecanone	7	3
2-Tridecanone	12,13	24
2-Pentadecanone	13	24
3-Hydroxy-2-butanone	12,13	24
2,3-Butanedione	12	24
2,3-Pentanedione	12	24
2,3-Octanedione	12	24
2-Furfuryl methyl ketone	14	25

TABLE 1 (continued)

Volatile Compounds Isolated from Heat-treated Beef

Compound	Meat sources and treatment*	References
Hydrocarbons		
Butane	5	9
Pentane	5	9
Hexane	5,7	3,9
Heptane	5	9
Octane	5	9
Nonane	12,13	24
Decane	12,13	24
Undecane	12,13	24
Dodecane	7,12,13	3,24
Tridecane	12,13	24
Tetradecane	12,13	24
Pentadecane	7,12,13	3,24
Hexadecane	7,12,13	3,24
Heptadecane	12,13	24
Octadecane	7,13	3,24
6(?) -Methyltetradecane	12	24
1-Pentadecene	7	3
1-Heptadecene	13	24
1-Undecene	7	3
Benzene compounds		
Benzene	7,8	3,12,13,14
Trimethylbenzene	12,13	24
Diethylbenzene	12	24
1,4-Dimethylbenzene	12,13	24
1,2-Dimethylbenzene	12	24
Propylbenzene	7	3
Butylbenzene	12	24
Benzaldehyde	4,7,8,12,13	3,12,13,14,24,29
3-Methylbenzaldehyde	7	3
Phenylacetaldehyde	8,13	12,13,14,24
Paradichlorobenzene	7	3
Toluene	7,12,13	3,24
Lactones		
δ -Valerolactone	13	24
γ -Valerolactone	7	3
γ -Butyrolactone	12,13	24
γ -Hexalactone	13	24
δ -Heptalactone	13	24
γ -Heptalactone	13	24
δ -Octalactone	13	24
γ -Octalactone	13	24
γ -Nonalactone	12,13	24
δ -Nonalactone	13	24
γ -Decalactone	13	24
Furans		
2-Acetylfuran	14	25
2-Pentylfuran	7,12,13	3,24
2-Hexylfuran	13	24
2-Heptylfuran	12,13	24
2-Octylfuran	13	24
2-Methyltetrahydrofuran-3-one	7	3
4-Hydroxy-2,5-dimethyl-3(2H)-furanone	4	28
4-Hydroxy-5-methyl-3(2H)-furanone	4	28

TABLE 1 (continued)

Volatile Compounds Isolated from Heat-treated Beef

Compound	Meat sources and treatment*	References
Sulfur compounds		
Hydrogen sulfide	1,2,3,7,9	4,5,6,7,11,15,16
Methyl mercaptan	1,2,4,7,10	4,5,8,11,15,16
Ethyl mercaptan	1,2,4,7	4,5,8,11,16
Propyl mercaptan	2	16
Butyl mercaptan	2,7	3,16
Isobutyl mercaptan	10	17
Dimethyl sulfide	1,4,7,9,10,12	3,4,5,8,15,16,17,24
Dimethyl disulfide	8,10	12,13,14,17
Ethyl methyl disulfide	10	17
Methyl vinyl disulfide	10	17
Diethyl disulfide	10	17
Methyl propyl sulfide	7	3
Methyl allyl sulfide	7	3
Diallyl sulfide	7	3
Thiophene	10	17
2-Methyl thiophene	10	17
2-Ethyl thiophene	10	17
2- <i>n</i> -Butyl thiophene	10	17
2- <i>n</i> -Amyl thiophene	10	17
<i>n</i> -Octyl thiophene	10	17
<i>n</i> -Tetradecyl thiophene	10	17
2-Thiophene carboxaldehyde	10	17
5-Methyl-2-thiophene carboxaldehyde	10	17
2,5-Dimethyl-3-thiophene carboxaldehyde	10	17
2-Thiophene acrolein	10	17
2-Acetyl thiophene	10	17
3-Acetyl thiophene	10	17
5-Methyl-2-acetylthiophene	10	17
1-(2-Thienyl)-1-propanone	10	17
1-(2-Methyl-5-thienyl)-1-propanone	10	17
2-Thiophene methanol	10	17
Tetrahydrothiophen-3-one	10	17
2-Methyl tetrahydrothiophen-3-one	10	17
Thiazole	10	17
2-Methylthiazole	10	17
4-Methylthiazole	10	17
2,4-Dimethylthiazole	10	17
5-Ethyl-4-methylthiazole	10	17
4-Ethyl-2-methylthiazole	10	17
2,4,5-Trimethylthiazole	10	17
2,4-Dimethyl-5-vinylthiazole	10	17
2-Acetylthiazole	10	17
Benzothiazole	10,14	17,25
3,5-Dimethyl-1,2,4-trithiolane	4,7,10	17,23,29
5,6-Dihydro-2,4,6-trimethyl-1,3,5-dithiazine	10	17
Trithioacetaldehyde	10	17
Trithioacetone	10	17
Methylthioacetate	10	17
1,1-bis-Methylthioethane	10	17
Dimethyl sulfone	12,13	24
2-Acetyl-2-thiazoline	4	27
1-Methylthio-ethanethiol	4	29
Thialdine	4	29

TABLE 1 (continued)

Volatile Compounds Isolated from Heat-treated Beef

Compound	Meat sources and treatment*	References
Nitrogen compounds		
Ammonia	3,9	6,7,15
Methylamine	3	6
2,4,5-Trimethyloxazoline	7	23
Pyrazine	11	18
Methylpyrazine	11,14	18,26
2,5-Dimethylpyrazine	11,14	18,26
2,6-Dimethylpyrazine	11,14	18,26
2,3-Dimethylpyrazine	11,14	18,26
Ethylpyrazine	11	18
2,5-Dimethyl-3-ethylpyrazine	14	26
2-Ethyl-6-methylpyrazine	11	18
2-Ethyl-5-methylpyrazine	11,14	18,26
Trimethylpyrazine	11,14	18,26
2,6-Diethyl-3-methylpyrazine	14	26
Tetramethylpyrazine	11,14	18,26
5-Ethyl-2,3-dimethylpyrazine	11	18
2-Ethyl-3,5-dimethylpyrazine	11	18
2-Ethyl-3,6-dimethylpyrazine	11	18
2,6-Diethylpyrazine	11	18
Methylpropylpyrazine	11	18
2-Methyl-3,5-diethylpyrazine	11	18
2-Methyl-3,6-diethylpyrazine	11	18
5-Methyl-2,3-diethylpyrazine	11	18
3,6-Diethyl-2,5-dimethylpyrazine	11	18
Triethylpyrazine	11	18
6,7-Dihydro-5(H)-cyclopentapyrazine	11	18
5-Methyl-6,7-dihydro-5(H)-cyclopenta- pyrazine	11	18
2(3),5-Dimethyl-6,7-dihydro-5(H)-cyclopenta- pyrazine	11	18
5,6,7,8-Tetrahydroquinoxaline	11	18
2-Methyl-5,6,7,8-tetrahydroquinoxaline	11	18
Vinylpyrazine	11	18
2-Methyl-6-vinylpyrazine	11	18
Isopropenylpyrazine	11	18
Acetylpyrazine	11	18
2-Methyl-5-acetylpyrazine	11	18
2-Ethyl-5-acetylpyrazine	11	18
1-Pyrazinyl-2-propanone	11	18

*Volatile compounds were isolated from the following sources:

1. Ox meat during boiling for meat extract.
2. Canned beef in broth.
3. Lyophilized lean muscle extract.
4. Boiled beef in broth.
5. Enzyme inactivated beef in cans.
6. Ground beef heated in water or fat.
7. Boiled beef.
8. Enzyme inactivated beef.
9. Boiled beef in slurry.
10. Pressure cooked beef (163° and 182°C) in slurry.
11. Pressure cooked beef (162.7°C) in slurry.
12. Roast beef heated to 163°C.
13. Roast beef drippings.
14. Shallow fried beef.

TABLE 2
Volatile Compounds from Processed Pork and Ham

Compound	Meat sources and treatment*	References
Acids		
Formic	2	20
Acetic	1,2	19,20
Propionic	2	20
Butyric	2	20
Isocaproic	2	20
Aldehydes		
Formaldehyde	2	20,22
Acetaldehyde	1,2,3,4	6,19,20,21,22
Propanal	1,2,3,4	6,19,20,21,22
Butanal	1,2,3	19,21,22,23
Pentanal	1,2	19,20,22
Hexanal	1,2,4	6,19,22
Heptanal	1,2	19,22
Octanal	1,2,4	6,19,22
Nonanal	1,2,4	6,19,22
Decanal	2	22
Undecanal	2,4	6,22
Dodecanal	1,2	19,22
Tridecanal	1	19
Tetradecanal	1	19
Pentadecanal	1	19
Hexadecanal	1	19
Heptadecanal	1	19
Octadecanal	1	19
Isobutyraldehyde	1,2,3	19,20,21
Isovaleraldehyde	1,2,3	19,20,21
2-Methylbutanal	1,2,3	19,20,21
2-Butenal	1	19
2-Methyl-2-butenal	1	19
2-Hexenal	2	22
2-Heptenal	1,2,4	6,19,22
2-Octenal	1,2,4	6,19,22
2-Nonenal	2,4	6,22
2-Decenal	2,4	6,22
2-Undecenal	2,4	6,22
2-Dodecenal	2	22
2,4-Heptadienal	2,4	6,22
2,4-Nonadienal	2,4	6,22
2,4-Decadienal	2,4	6,22
2,4-Undecadienal	2	22
2,4-Dodecadienal	2	22
2-Phenyl-2-butenal	1	19
5-Methyl-2-phenyl-2-hexenal	1	19
Benzaldehyde	1	19
Phenylacetaldehyde	1	19
Ketones		
Acetone	1,2,3,4	6,19,20,21
2-Butanone	1,2	19,20
Diacetyl	2	20
2-Heptanone	1	19
2-Octanone	1	19
2-Decanone	1	19

TABLE 2 (continued)

Volatile Compounds from Processed Pork and Ham

Compound	Meat sources and treatment*	References
Ketones (cont.)		
2-Tridecanone	1	19
2-Heptadecanone	1	19
3-Pentanone	1	19
3-Octanone	1	19
4-Methyl-2-pentanone	1	19
3-Buten-2-one	1	19
3-Methyl-3-buten-2-one	1	19
3-Penten-2-one	1	19
2,3-Butanedione	1	19
2,3-Pentanedione	1	19
Cyclopentanone	1	19
2-Cyclopenten-1-one	1	19
1-Phenyl-2-propanone	1	19
1-Hydroxy-2-propanone	1	19
3-Hydroxy-2-butanone	1	19
3-Hydroxy-2-pentanone	1	19
2-Hydroxy-3-pentanone	1	19
Esters		
Methyl formate	1	19
Ethyl formate	1	19
Ethyl acetate	1	19
Pentyl acetate	1	19
Acetyl acetate	1	19
Ethyl propionate	1	19
Ethyl isovalerate	1	19
Ethyl dodecanoate	1	19
Ethyl tetradecanoate	1	19
Methyl hexadecanoate	1	19
Alcohols		
Methanol	1	19
Ethanol	1	19
2-Propanol	1	19
Butanol	1	19
2-Methyl-1-propanol	1	19
2-Butanol	1	19
3-Methylbutanol	1	19
2-Methyl-2-butanol	1	19
2-Methyl-3-buten-2-ol	1	19
3-Octenol	1	19
Hydrocarbons		
2-Pentene	1	19
Heptane	1	19
Pentadecene	1	19
Pentadecane	1	19
Hexadecene	1	19
Hexadecane	1	19
Heptadecane	1	19
Benzene	1	19
Ethylbenzene	1	19
Hexylbenzene	1	19
Toluene	1	19
m-Xylene	1	19
Limonene	1	19

TABLE 2 (continued)

Volatile Compounds from Processed Pork and Ham

Compound	Meat sources and treatment*	References
Lactones		
δ-Nonalactone	1	19
δ-Decalactone	1	19
Furans		
2-Methylfuran	1	19
2-Ethylfuran	1	19
2-Butylfuran	1	19
2-Pentylfuran	1	19
2-Furaldehyde	1	19
5-Methyl-2-furaldehyde	1	19
Furfuryl alcohol	1	19
2-Acetylfuran	1	19
Propionylfuran	1	19
Furfuryl methyl ketone	1	19
2-Methyltetrahydrofuran-3-one	1	19
Furfuryl formate	1	19
Furfuryl acetate	1	19
Furfuryl propionate	1	19
Furfuryl butyrate	1	19
Furfuryl pentanoate	1	19
Furfuryl hexanoate	1	19
Ethyl furoate	1	19
Ethyl furfuryl ether	1	19
Furfuryl ether	1	19
2,2'-Methylenedifuran	1	19
5-Methyl-2,2'-methylenedifuran	1	19
2(or 3)-Phenylfuran	1	19
Thiazoles		
Thiazole	1	19
4-Methylthiazole	1	19
2-Acetylthiazole	1	19
Thiophenes		
2-Methylthiophene	1	19
2-Thiophenecarboxaldehyde	1	19
3-Thiophenecarboxaldehyde	1	19
3-Methyl-2-thiophenecarboxaldehyde	1	19
5-Methyl-2-thiophenecarboxaldehyde	1	19
2,5-Dimethyl-3-thiophenecarboxaldehyde	1	19
2-Acetylthiophene	1	19
3-Acetylthiophene	1	19
5-Methyl-2-acetylthiophene	1	19
Thiopheneacrolein	1	19
Sulfur compounds		
Hydrogen sulfide	2,3	21
Methanethiol	1,2,3	19,21
Methyl sulfide	1	19
Methyl disulfide	1	19
Methyl sulfone	1	19
Methyl sulfoxide	1	19
Methyl thioacetate	1	19
Methyl thiopropionate	1	19

TABLE 2 (continued)

Volatile Compounds from Processed Pork and Ham

Compound	Meat sources and treatment*	References
Sulfur compounds (cont.)		
Ethyl thiopropionate	1	19
Benzyl methyl sulfide	1	19
Furfuryl methyl sulfide	1	19
Furfuryl methyl disulfide	1	19
Pyrroles		
Pyrrole-2-carboxaldehyde	1	19
5-Methylpyrrole-2-carboxaldehyde	1	19
Acetylpyrrole	1	19
2-Acetylpyrrole	1	19
2-Propionylpyrrole	1	19
Pyrazines		
Pyrazine	1	19
Methylpyrazine	1	19
Ethylpyrazine	1	19
2,3-Dimethylpyrazine	1	19
2,5-Diethylpyrazine	1	19
2,6-Dimethylpyrazine	1	19
2-Ethyl-5-methylpyrazine	1	19
2-Ethyl-6-methylpyrazine	1	19
2,5-Diethylpyrazine	1	19
Trimethylpyrazine	1	19
2-Ethyl-3,5-dimethylpyrazine	1	19
2-Ethyl-3,6-dimethylpyrazine	1	19
5-Ethyl-2,3-dimethylpyrazine	1	19
2,3-Diethyl-5-methylpyrazine	1	19
2,5-Diethyl-3-methylpyrazine	1	19
3,5-Diethyl-2-methylpyrazine	1	19
Triethylpyrazine	1	19
Tetramethylpyrazine	1	19
2-Methyl-3-vinylpyrazine	1	19
2-Methyl-5-vinylpyrazine	1	19
Acetylpyrazine	1	19
2-Methyl-5(or 6)-acetylpyrazine	1	19
2-Ethyl-3-acetylpyrazine	1	19
2-Ethyl-5(or 6)-acetylpyrazine	1	19
6,7-Dihydro-5H-cyclopentapyrazine	1	19
2-Methyl-6,7-dihydro-5H-cyclopentapyrazine	1	19
5-Methyl-6,7-dihydro-5H-cyclopentapyrazine	1	19
2(or 3),5-Dimethyl-6,7-dihydro-5H-cyclopentapyrazine	1	19
Quinoxaline	1	19
2-Methylquinoxaline	1	19
6-Methylquinoxaline	1	19
5,6,7,8-Tetrahydroquinoxaline	1	19
2-Methyl-5,6,7,8-tetrahydroquinoxaline	1	19
(2-Furyl)pyrazine	1	19
2-(2-Furyl)-5(or 6)-methylpyrazine	1	19

TABLE 2 (continued)

Volatile Compounds from Processed Pork and Ham

Compound	Meat sources and treatment*	References
Miscellaneous compounds		
Ethyl vinyl ether	1	19
Diethyl ether	1	19
Acetaldehyde, diethyl acetal	1	19
Phenol	1	19
Trimethyloxazole	1	19
Vanillonitrile	1	19

*Volatile compounds were isolated from the following sources:

1. Pressure cooked (157°C) pork liver.
2. Dry cured ham.
3. Uncured ham.
4. Pork lipids heated at 100°C.

2. Relatively less volatile fraction having a pleasant meaty aroma. Lactic acid and its ammonium salt were the only compounds identified in this fraction.

Of the large number of volatile compounds identified in meat products, Herz and Chang³⁰ believe that lactones, furan ring compounds that do not contain sulfur, and aliphatic sulfur compounds make a direct contribution to the meat flavor profile. They, however, concede that none of the representatives of the above classes of compounds isolated from cooked meat by their group^{3,23,30} has a characteristic meaty aroma. On the other hand, these researchers have reported the presence of four volatile compounds in cooked beef that have a meaty aroma.^{3,23} These compounds include: 5-thiomethylfurfural, thiophencarboxy-2-aldehyde, 2,5-dimethyl-1,3,4-trithiolane, and 2,4,5-trimethyl-3-oxazoline.

Watanabe and Sato²⁶ reported that in shallow fried beef, the most likely compounds responsible for the typical roasted flavor are alkyl-substituted pyrazines and pyridines. This deduction was based on their observation that the removal of basic compounds from the total flavor extract in diethyl ether with hydrochloric acid solution resulted in a considerable loss of the roasted flavor. In a subsequent study with shallow fried beef, Watanabe and Sato²⁵ isolated a volatile fraction with a typical heated beef flavor and observed that no single compound isolated in this fraction had a typical heated beef flavor. It was suggested that the heated beef flavor might be a complex sensation resulting from a mixture of several different compounds: methional, 2-acetylfuran, 2-furfuryl methyl ketone, 1-methyl-2-acetylpyrrole, benzothiazole, and others.

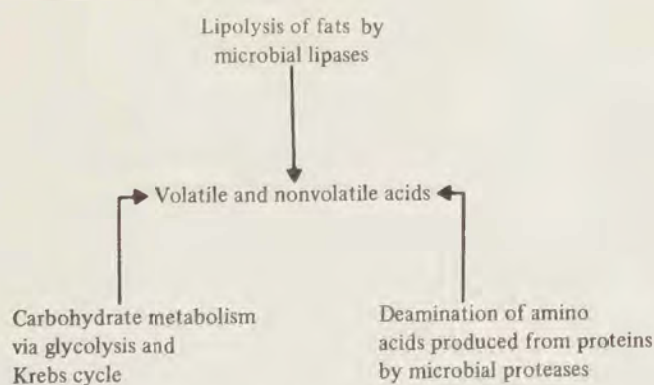
It appears likely that the question of the nature of compounds responsible for the characteristic cooked meat flavor would remain unresolved for some time to come. It may, however, be pointed out that most of the odorous compounds present in cooked meat are likely to play a significant role in determining its flavor character. It is also common knowledge that odorous compounds, when presented in a mixture, often give an odor sensation somewhat different from the one predicted on the basis of odor characteristics of the individual components in the mixture. It may, therefore, be futile to look for the "characteristic" compounds responsible for the typical meat flavor. Most likely, the subtle quantitative balance of the various odorous components in cooked meat determines the aroma character of meat. For example, the aromas of cooked beef and cooked pork liver are easily distinguishable, although a cursory look at the volatile compounds identified in them (Tables 1 and 2) shows a striking similarity.

Unlike the cooked meat aroma produced from their precursors during heat treatment, the characteristic aroma of dry or fermented sausages is related, in part, to the hydrolytic and oxidative changes occurring in the lipid fraction during ripening.^{31,32} The bacterial,³³⁻³⁵ muscle, and adipose tissue³⁶ lipases liberate fatty acids from the sausage lipids. The unsaturated fatty acids thus liberated undergo nonenzymatic

oxidation and produce lipid peroxides and carbonyl compounds.^{3,1,4,7} The lipid peroxides are probably further metabolized by the sausage microflora to carbonyl compounds and fatty acids.^{3,8,3,9} Demeyer et al.^{4,0} have recently investigated the specificity of lipolysis during dry sausage ripening and report that unsaturated fatty acids are selectively liberated by the lipases from the sausage lipids. In this study, linoleic acid was liberated into the free fatty acid fraction at a much faster rate than all other acids. The rate of lipolysis decreased in the order: linoleic > oleic > stearic > palmitic acid. Since pork fat is known to have most of the linoleic and oleic acids (50 to 60%) at position 3 of triglyceride molecule,^{4,1} it has been suggested by Demeyer and co-workers^{4,0} that the lipases occurring in sausages have specificity for position 3 of triglycerides. The difference observed for linoleic and oleic acids may be related to specificity of the enzyme for the fatty acid structure, as both positional and structural specificity is known to occur in microbial lipases.^{4,1} Since only unsaturated fatty acids undergo nonenzymatic oxidation and produce lipid peroxides and carbonyl compounds, the specificity of microbial lipases for position 3 of triglycerides, which in the case of pork lipids contains a greater percentage of unsaturated fatty acids than positions 1 and 2 of triglyceride molecule, appears to be an important factor in the development of ripened sausage flavor. Micrococci^{3,2,3,3} and to a lesser degree lactobacilli^{4,2,4,3} have been reported to be responsible for lipolysis in dry sausage.

During sausage ripening, the carbonyl concentration of the product increases severalfold. In a study carried out by Demeyer et al.,^{4,0} carbonyl compounds increased in the sausage during the first week of ripening, decreased after smoking, and again increased to final values of about 300 $\mu\text{mol}/100\text{ g}$ of dry matter. The initial increase was attributed to compounds formed in carbohydrate fermentation,^{4,4} while the increase in the last stages might have been due to further metabolism of lipid peroxides.^{3,9}

The volatile fatty acids produced by the microorganisms during sausage ripening are very important in the dry sausage aroma. Significant concentrations of acetic, propionic, and butyric acids have been reported in fermented sausages, along with large amounts of nonvolatile lactic acid which affects the taste and reduces the pH of sausage from an average of 5.8 to 4.8.^{4,4} The homo- and hetero-fermentative metabolic pathways, which may be involved in the formation of volatile fatty acids and carbonyl compounds by the microorganisms during dry sausage ripening, were discussed in detail in the earlier review.² Among the volatile fatty acids mentioned above, acetic acid may be formed from aspartic acid, alanine, serine, glycine, cystine, threonine, and glutamic acid by the bacterial metabolism. Propionic acid may originate from alanine and serine. Butyric acid may be formed from threonine and glutamic acid. These fatty acids may also be synthesized from fats and carbohydrates as shown below:



Nonvolatile Flavor Compounds and Flavor Potentiators

The overall composition of meat extracts and the possible taste effect of various compounds identified have been extensively investigated.^{4,5-6,2} Inosine-5'-monophosphate (IMP) is a major component of meat extracts. It is considered a key flavor component responsible for the strong meaty taste.^{5,8,6,3,6,4} Other nonvolatile components with taste properties include amino acids, peptides, and other nucleotides. The high content of IMP in meat extracts is due to the presence of adenosine-5'-triphosphate (ATP) as a major mononucleotide in the muscle of living animals. After slaughter there is a rapid transformation of this

nucleotide to adenosine-5'-monophosphate (AMP), which is then deaminated to IMP.⁶⁴ A partial list of compounds isolated from meat extracts is presented in Table 3. Several of the compounds listed in this table act as flavor precursors. The meat flavor precursors are discussed in detail later in this article. The contribution of peptides and amino acids to the taste of foodstuffs has been investigated by Kirimura and co-workers.⁶⁵ The organoleptic characteristics of amino acids reported by these workers are reproduced in Table 4.

TABLE 3

Potential Flavor Precursors Isolated from Meat Extracts

Alanine	Fructose-6'-phosphate	Lysine
β -Alanine	Glucose	Methionine
quaternary Amines	Glucose-6'-phosphate	Methylhistidine
Ammonia	Glutamic acid	Nicotinamide-adeninedinucleotide
Anserine	Glutamine	Ornithine
Arginine	Glutathione	Peptides
Asparagine	Glycerophosphoethanolamine	Phenylalanine
Aspartic acid	Glycine	Phosphoethanolamine
Carnitine	Glycoproteins	Phosphoserine
Carnosine	Histidine	Proline
Citrulline	Hydroxyproline	Purine-nucleosides
Creatine	Hypoxanthine	Purine-nucleotides
Creatinine	Inosine-5'-monophosphate	Ribose
Cysteine	and other nucleotides	Ribose-5'-phosphate
Cystine	Isoleucine	Serine
Fructose	Leucine	Taurine
		Threonine
		Tyrosine
		Urea
		Valine

TABLE 4

Organoleptic Characteristics of Amino Acids

Amino acid	Relative taste intensities ^a				
	Sweet	Salty	Sour	Bitter	MSG-like
Sweet amino acids					
Hydroxyproline	***			**	
Lysine·HCl	**			**	
Alanine	***				
Glycine	***				
Serine	***		*		*
Glutamine	*				*
Threonine	***		*	*	
Proline	***			***	
Sour and MSG-like amino acids					
Aspartic acid			***		*
Glutamic acid			***		**
Histidine·HCl			***		
Asparagine			**	*	
Monosodium glutamate(MSG)	*	*			***
Na-aspartate		**			**

TABLE 4 (continued)

Organoleptic Characteristics of Amino Acids

Amino acid	Relative taste intensities ^a				
	Sweet	Salty	Sour	Bitter	MSG-like
Bitter amino acids					
Histidine				**	
Arginine-HCl				***	
Methionine				***	*
Valine				***	
Arginine				***	
Isoleucine				***	
Phenylalanine				***	
Tryptophan				***	
Leucine				***	

^aAsterisks denote relative taste intensities.

From Kirimura, J., Shimizu, A., Kimizuka, A., Ninomiya, T., and Katsuya, N., *J. Agric. Food Chem.*, 17, 689, 1969. With permission.

ORIGIN OF MEAT FLAVOR

Patents on Synthetic Meat Flavors

The importance of the flavor precursors, listed in Table 3, in the formation of meat flavor may be easily recognized by the fact that a large number of patents on meat flavoring compositions, which are based on the heat treatment of flavor precursors, have been awarded in the last few years. Some of the patents are based on the Maillard reactions between sugars and amino compounds. Other make use of the strong flavor potentiation of nucleotide derivatives, along with sugar-amine interactions. A sample of the important patents issued on synthetic meat flavors, in the last few years, is summarized below:

A patent issued to Unilever⁶⁶ in 1966 details a method of preparing flavoring compounds by heating a mixture of carbohydrates containing at least one amino acid in the presence of a fatty acid. The carbohydrate may be a pentose or a hexose or a polysaccharide, or a mixture of these. There may be a single amino acid or a mixture of two or more amino acids. For example, a mixture of 480 g wheat gluten hydrolyzate, 56 g cysteine-HCl, 70 g xylose, 50 g oleic acid, and 1,500 g water was heated for 2 hr with stirring in an oil bath at 120°C, then cooled to 50°C, and a slurry of 400 g cornmeal in 600 g water was added with vigorous stirring. A yellowish brown powder was obtained that had the flavor and aroma of roast mutton when dispersed in a small quantity of water.

A similar Unilever patent⁶⁷ deals with mixtures of amino acids including glutamic acid, nucleotides (preferably GMP and IMP), succinic acid, and hydroxycarboxylic acids such as lactic acid in a defined ratio to have more pronounced meaty flavor.

Another Unilever patent⁶⁸ is concerned with the meat flavorings containing at least one of the optical antipodes of pyrrolidonecarboxylic acid. In flavorings also containing other additives, the preferred weight ratios were: pyrrolidonecarboxylic acid/glutamine/succinic acid/5'-ribonucleotides = 1/(0-25)/(0-1)/(0-5).

In 1970, Van der Ouweland and Peer⁶⁹ were awarded a patent, assigned to Unilever, on a synthetic meat flavor utilizing the capability of certain heterocyclic ketones such as 4-hydroxy-5-methyl-2,3-dihydrofuran-3-one, to react with inorganic or organic sulfur compounds, and to form compounds with an aroma similar to that of cooked or roast meat.

A recent patent awarded to Brinkman and Van der Heyden,⁷⁰ assigned to Lever Brothers Co., claims that the addition of thiaalkanethiols, where two aliphatic groups are methyl- or ethyl-radical, or their salts,

in an amount such that the food contains 0.1 to 1,000 mg compound/1,000 kg product results in a rich meat flavor. 2-Thiabutane-3-thiol was cited as an example of thiaalkanethiol in the patent.

Kyowa Fermentation Industry was awarded a patent on meat flavoring composition⁷¹ prepared by applying the Maillard reaction to ribose phosphate and an amino acid, such as lysine, hydrolyzed protein, or a casein hydrolyzate at 150 to 180°C.

Kitada et al.⁷² were awarded a patent, assigned to Ajinomoto Co., on a Maillard reaction product having a meat-like flavor. Meat flavor was produced by heating a solution containing cystine and/or cysteine and a reducing sugar at 50 to 120°C at a pressure of 70 to 200 kg/cm².

Another Ajinomoto patent⁷³ discloses the synthetic compounds which enhance the meat flavor. Such compounds were obtained by mixing amino acids, organic acids, phosphates, 5'-nucleotides, and analogous compounds in water, adjusting pH of the solution to about 5 and then cooking it for a few minutes.

International Flavors & Fragrances patented a flavoring material⁷⁴ with a strong meat flavor by reacting thiamine or 3-acetyl-3-mercaptopropanol-1 in the presence of an organic acid.

Giacino patent,⁷⁵ assigned to International Flavors & Fragrances, discloses the preparation of meat-like flavors by heating a mixture comprised of taurine and thiamine and then adding it to a source of free amino acids.

Thomas patent,⁷⁶ assigned to Pfizer Inc., discloses synthetic ham- and bacon-flavored compositions prepared by heating an aqueous mixture of hexose or pentose, cystine or cysteine, and glycine. After cooling the mixture, monosodium glutamate, protein hydrolyzate, sucrose, edible fats, ribonucleotide, and hickory smoke flavor are incorporated to obtain the desired flavor.

Broderick and Marcus patent,⁷⁷ assigned to Kohnstamm and Co., discloses a procedure of preparing artificial flavoring substances with the aroma and taste of cooked meat by causing a ribose moiety-containing ribonucleotide to react at elevated temperature with cystine or cysteine in the presence of water.

The use of thiamine as one of the ingredients used in the preparation of synthetic meat flavors is not surprising. Thiamine, like cystine and cysteine, on heating produces a number of sulfur compounds that contribute significantly in meat-like synthetic flavor formulations. It should be noted here that meat flavor being a very characteristic and complex flavor has not yet lent itself to synthetic duplication. The patents on synthetic meat flavors discussed above and the others may be regarded as extenders of meat flavor, rather than the true meat flavor. However, the synthetic preparation of the characteristic flavors of different cuts of meat and species flavor, such as pork, lamb, and beef flavor, are within the realm of being feasible.

The Role of Enzymes in the Development of Flavor Precursors

The nature and concentration of meat flavor precursors depends, to a large extent, on the metabolic activities in the animal body and the post-mortem storage conditions. The biochemical reactions in a living system are catalyzed by enzymes. The nature and concentration of the enzymes found in different parts of animal body are directly responsible for the accumulation or depletion of metabolites, some of which act as flavor precursors, in muscle. The enzymatic reactions as related to the development of flavor precursors have not been studied to any significant extent. However, a considerable amount of indirect evidence is available. This evidence is presented under the headings: ante-mortem and post-mortem factors.

Ante-mortem Factors

The following observations suggest the importance of enzymes in the development of flavor precursors:

Age of the animal at the time of slaughter is related to cooked meat flavor. Barbella et al.⁷⁸ and more recently Lowe and Kastelic⁷⁹ reported that young calves up to about 11 months lack the typical flavor quality and intensity of beef. The change in metabolism with age is well known and it is an important factor in the development of a normal meat flavor. The change in the composition of muscle with age is another important factor. It is safe to assume that if flavor is related to the concentration of metabolites in muscle, an increase in intensity and a change in profile up to an age of a little metabolic change should be

expected. A similar observation was made by Crocker⁸⁰ who reported that odor of cooked meat generally increases with age of the animal, as the distinctive taste of beef and insipid taste of veal testify. It is well known that myoglobin content of muscle increases with age. However, this increase is not likely to influence the flavor. It is more likely that this difference may be due to an increase in the concentration of certain amino acids; for example, Gruhm⁸¹ has reported that the relative concentration of valine, methionine, leucine, and isoleucine increases with age, or to some constituents of intramuscular fat which alter with increasing maturity.

An interesting study made on 10 pairs of bull and steer twins by Bryce-Jones et al.⁸² showed that steers were definitely more tender and also better than bulls in juiciness and flavor.

The effect of breed on flavor is less widely recognized, but scientific studies have revealed that this factor certainly has an influence. European-type cattle give a better flavored meat than do Brahmin cattle,⁸³ though it is not clear to what extent this difference may be affected by the undoubted toughness of the latter. Again, the introduction of Hampshire blood into a large white pig stock has been reported to favor an enhanced tendency to rancidity and thus to an off flavor development.

The sex of animal also influences the flavor of meat to some extent. According to Weir⁸⁴ mature males have a "staggy" odor. It is probably in regard to off flavor that the influence of the sex of an animal is most clearly noted. Thus, male pigs at maturity are frequently associated with an unpleasant "boar odor" on cooking. Patterson^{85,86} has recently shown that the substance responsible is probably 5-androst-16-ene-3-one and, what is particularly interesting, this is closely related to a corresponding alcohol in the salivary gland of mature boar which acts as a stimulant to female at the time of mating.

Differences in meat flavor arising from the type of joint are widely recognized. Thus, the rump and topside yield roasts with markedly greater flavor though also greater toughness than those from the sirloin.⁵⁴

Electric shock treatment during the 24 hr preceding slaughter at 20 min intervals invariably increases the pH of cooked steaks from Angus or Hereford steers, compared with control steaks from paired animals receiving no shock treatment.⁸⁷

Feeding of antibiotics or tranquilizers could affect meat flavor. The effect on rumen or intestinal flora may be important in the case of antibiotics, because the digestion of ruminants relies on the action of bacteria, and the fatty acids produced by these microorganisms can modify the lipids of the animal. Harris et al.⁸⁸ reported that meat from conventionally raised chicken had a more characteristic flavor than meat from the chicken born and raised in a germfree environment.

A relatively high pH results from injecting adrenaline just ante-mortem.⁸⁹ Ante-mortem feeding of sucrose, on the other hand, lowers pH and increases lactate content of pork from pigs that had been transported 340 mi in 24 hr prior to slaughtering; the same effect was not noticed in animals transported only 3.1 mi.⁹⁰

Proteolytic enzymes have been successfully injected ante-mortem into the bloodstream in order to tenderize the meat. Although no qualitative differences in lower boiling aroma volatiles of beef steak from the animal injected with papain were observed, some differences in quantities of the individual compounds were noticed.⁹¹

Biochemical state of the muscle is also important. Post-mortem all muscles convert their stores of glycogen into lactic acid causing the pH to fall to an ultimate value of about 5.5. If glycogen store is lower than 0.6% of the muscle weight, the extent of this post-mortem glycolysis results in an ultimate pH value between 5.5 and 7. A relationship between the ultimate pH of muscle and its flavor has been reported.⁹²

Let us now examine how the flavor precursors are produced in an animal body. It is quite clear from the patent literature cited earlier and other reported findings that meat flavor precursors include amino acids, small peptides, carbohydrates, particularly hexoses and pentoses, and nucleotides (Table 3). As illustrated in Figure 1, amino acids found in blood and tissues may result from their absorption from intestines, breakdown of tissue proteins, and synthesis of amino acids, particularly in the liver. These amino acids are used by the animal body in a number of ways. The major pathways and products of amino acid metabolism are given in Table 5. Considering various metabolic functions of different animal tissues, it appears likely that most of the amino acids found in meat are produced by the breakdown of tissue proteins. In that case,

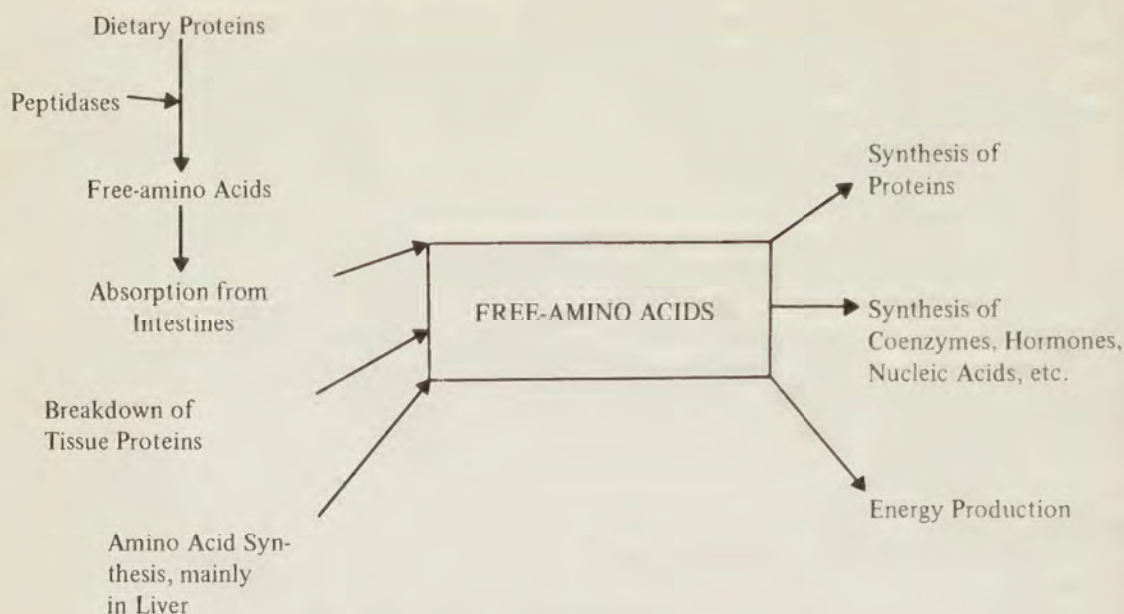


FIGURE 1. Fate of amino acids in mammalian body.

the nature and concentration of amino acids in the muscle would depend on the genetic make up and physiological state of the animal at the time of slaughter. Post-mortem, tissue proteins continue to degrade and release amino acids by the action of native muscle and microbial proteases.

Other metabolic pathways which may have a role in the development of meat flavor precursors are given in Figures 2 to 8.

Figure 2 refers to the mammalian fatty acid metabolism. The compounds isolated from meat extracts, which may originate from one or more of the metabolic schemes described, are acetoin, diacetyl, formic acid, acetic acid, hydroxybutyric acid, and succinic acid. It is widely recognized that the species flavor of meat develops from adipose tissues. The depot fat and water soluble constituents of adipose tissues probably interact to produce the species flavor. The nature of depot fat is governed by the genetic make up and diet of the animal. The various pathways involving fatty acid, glycerolipid, sphingolipid, and sterol metabolism, which are called upon by the living system and influence the make up of depot fat, are outlined in Figures 2 to 5. The role of depot lipids in the development of meat flavor is discussed later in this review.

The carbohydrates present in raw meat interact with amino compounds and produce volatile flavor compounds via Maillard reactions. These reactions are discussed in detail in a later section in this article. Sugars and sugar phosphates identified from meat extracts include: fructose, fructose-6'-phosphate, fructose-1,6'-diphosphate, glucose, glucose-6'-phosphate, ribose, and ribose-5'-phosphate (Table 3). The pathways of carbohydrate metabolism and interrelationship of sugars and sugar phosphates discussed above, are shown in Figure 6. These metabolic schemes also depict the synthesis of lactic acid from glycogen of the muscle. The amount of lactic acid present in meat is important in its characteristic taste.

Figure 7 gives the metabolic interrelationship of lipids, carbohydrates, and proteins. These metabolic schemes show how a metabolite such as acetate may originate from proteins, lipids, or carbohydrates.

Figure 8 details pathways of nucleoprotein catabolism. Nucleoproteins are composed of basic proteins (histones or protamines) associated with nucleic acids in a salt linkage. The reactions presented are for ribonucleic acid (RNA). A parallel series occurs with deoxyribonucleic acid (DNA). The breakdown products of nucleic acids, produced enzymatically post-mortem, are very important in meat flavor, particularly inosine 5'-monophosphate. The specific reactions involved in the formation of this important nucleotide are discussed in detail in a later section.

TABLE 5

Pathways of Amino Acid Metabolism

Amino acid	Product of oxidative deamination or transamination	Product of decarboxylation	Pathways and products of metabolism
1. L-Alanine	Pyruvic acid		
2. L-Arginine	α -Keto- δ -guanidovaleric acid	Agmatine	Arginine $\rightarrow$ ornithine + urea; arginine $\rightarrow$ citrulline + NH_3 ; arginine + glycine $\leftrightarrow$ guanidoacetic acid + ornithine
3. L-Asparagine	α -Ketosuccinamic acid		Asparagine $\rightleftharpoons$ aspartic acid + NH_3 ; α -ketosuccinamic acid $\rightarrow$ NH_3 + oxaloacetic acid
4. L-Aspartic acid	Oxaloacetic acid	α -Alanine	Aspartic acid + carbamylphosphate $\rightarrow$ P_i + carbamylaspartic acid $\rightarrow$ $\rightarrow$ pyrimidines; aspartic acid $\leftrightarrow$ fumaric acid + NH_3 ; aspartic acid $\rightarrow$ aspartic semialdehyde $\rightarrow$ homoserine $\rightarrow$ (i) threonine, (ii) methionine, or (iii) lysine; aspartic acid $\rightarrow$ nitrogen of purine ring (<i>see</i> purine metabolism); aspartic acid + IMP $\rightarrow$ adenylosuccinate $\rightarrow$ AMP + fumarate. <i>See also</i> asparagine.
5. L-Citrulline	α -Keto- δ -carbamidovaleric acid		Citrulline + aspartic acid $\rightarrow$ argininosuccinic acid $\leftrightarrow$ arginine + fumaric acid; citrulline + $\text{P}_i \leftrightarrow$ ornithine + carbamylphosphate; carbamylphosphate + ADP $\leftrightarrow$ CO_2 + NH_3 + ATP
6. L-Cysteine and L-cystine	β -Mercaptopyruvic acid		Cysteine $\rightarrow$ H_2S + NH_3 + pyruvic acid; β -mercaptopyruvic acid $\rightarrow$ pyruvic acid + S ; cysteine $\rightarrow$ cysteine sulfinic acid $\rightarrow$ (i) cysteic acid $\rightarrow$ CO_2 + taurine, (ii) CO_2 + hypotaurine, or (iii) via transamination $\rightarrow$ β -sulfinylpyruvate $\rightarrow$ pyruvate + SO_3^{2-} ; 2 cysteine $\leftrightarrow$ cystine. <i>See also</i> methionine for biosynthesis of cysteine.
7. L-Glutamic acid	α -Ketoglutaric acid	γ -Aminobutyric acid	Glutamic acid $\leftrightarrow$ glutamic semialdehyde $\leftrightarrow$ (i) ornithine or (ii) Δ^1 -pyrroline-5-carboxylate $\leftrightarrow$ proline; glutamic acid $\rightarrow$ <i>N</i> -acetylglutamic acid $\leftrightarrow$ <i>N</i> -acetylglutamic semialdehyde $\leftrightarrow$ <i>N</i> -acetylornithine $\rightarrow$ ornithine. <i>See also</i> glutamine, histidine.
8. L-Glutamine	α -Ketoglutaric acid		α -Ketoglutaric acid $\rightarrow$ NH_3 + α -ketoglutarate; glutamine $\rightleftharpoons$ NH_3 + glutamic acid; glutamine + 5'-phospho- α -D-ribose/pyrophosphate $\rightarrow$ glutamate + 5'-phospho- β -D-ribose/lysine $\rightarrow$ $\rightarrow$ 5'-phosphoribosyl- <i>N</i> -formylglycineamide + glutamine $\rightarrow$ glutamate + 5'-phosphoribosyl- <i>N</i> -formylglycineamide $\rightarrow$ $\rightarrow$ purines; glutamine + xanthylic acid $\rightarrow$ guanylic acid + glutamate; glutamine + fructose-6-phosphate $\rightarrow$ glucosamine-6-phosphate + glutamate. <i>See also</i> histidine.

Pathways of Amino Acid Metabolism

Amino acid	Product of oxidative deamination or transamination	Product of decarboxylation
9. Glycine	Glyoxylic acid	5,10-Methylene tetrahydrofolate + NH ₃ + 2H
10. L-Histidine	β -Imidazolepyruvic acid	Histamine
11. Hydroxyproline	α -Keto-2-hydroxy- δ -aminovaleric acid	
12. L-Isoleucine	α -Keto- β -methylvaleric acid	2-Methylbutylamine
13. L-Leucine	α -Ketoisocaproic acid	3-Methylbutylamine
14. L-Lysine	α -Keto- ϵ -aminocaproic acid	Cadaverine

TABLE 5 (continued)

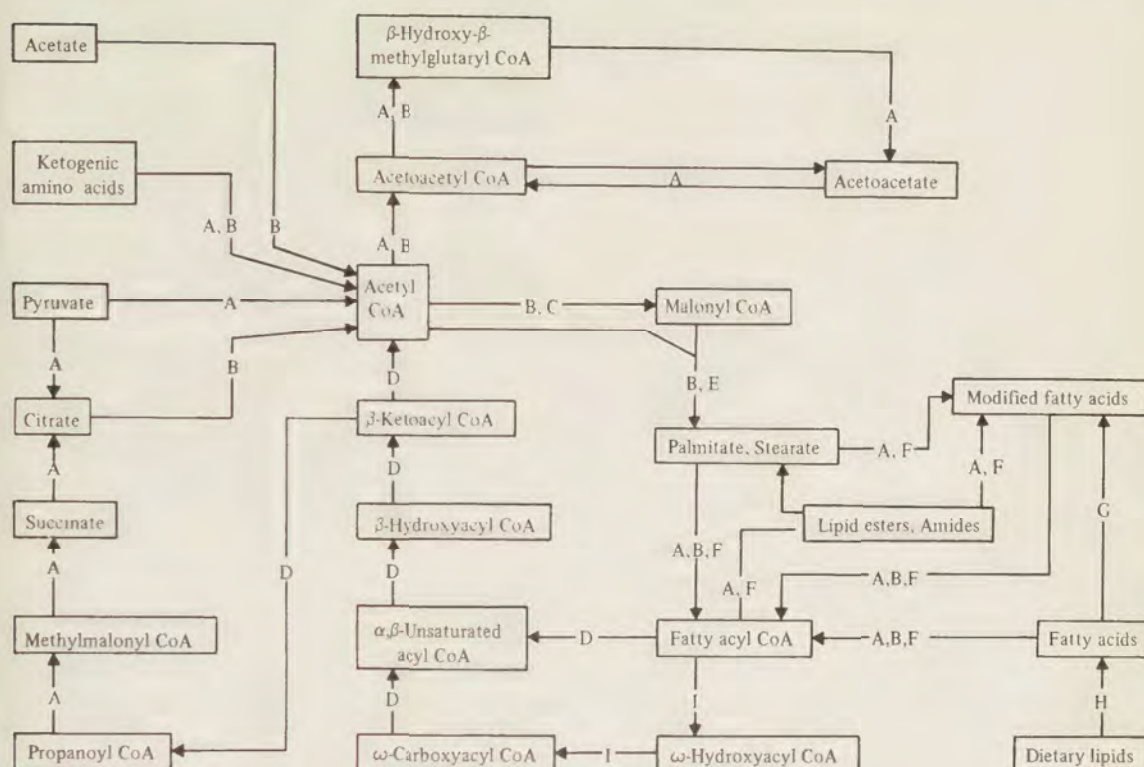
Pathways of Amino Acid Metabolism			Product of oxidative deamination or transamination	Product of decarboxylation	Pathways and products of metabolism
Amino acid					
15. L-Methionine		α -Keto- γ -methylbutyric acid			Methionine + ATP $\rightarrow$ PP _i + P _i + S-adenosylmethionine (active methyl donor); S-adenosylmethionine + guanidoacetate (<i>see</i> arginine) $\rightarrow$ creatine + S-adenosylhomocysteine; S-adenosylhomocysteine $\rightarrow$ adenosine + homocysteine; homocysteine + serine $\rightarrow$ cystathionine $\rightarrow$ cysteine + homoserine; homoserine $\rightarrow$ α -ketobutyrate + NH ₃ ; α -ketobutyrate $\rightarrow$ CO ₂ + propionyl CoA $\rightarrow$ methylmalonyl CoA $\rightarrow$ succinyl CoA; homoserine (<i>see</i> aspartic acid) + cysteine $\rightarrow$ cystathionine $\rightarrow$ serine + homocysteine; homocysteine + one C donor $\rightarrow$ methionine; homocysteine $\rightarrow$ (i) homocysteic acid or (ii) NH ₃ + H ₂ S + α -ketobutyrate.
16. L-Ornithine		Glutamic semialdehyde, or α -keto- δ -aminovaleric acid		Putrescine	Ornithine $\leftrightarrow$ proline; ornithine $\leftrightarrow$ glutamic semialdehyde $\leftrightarrow$ glutamate; ornithine + carbamylphosphate $\rightarrow$ citrulline. <i>See also</i> citrulline, glutamic acid.
17. L-Phenylalanine		Phenylpyruvic acid		Phenylethylamine	Phenylalanine $\rightarrow$ tyrosine; phenylpyruvic acid $\rightarrow$ (i) phenyllactic acid or (ii) CO ₂ + phenylacetic acid; phosphoenolpyruvate + erythrose-4-phosphate $\rightarrow$ 3-deoxy-D-arabino-heptulosonic acid-7-phosphate $\rightarrow$ $\rightarrow$ 5-dehydroquininate $\rightarrow$ 5-dehydroshikimate $\rightarrow$ shikimate $\rightarrow$ shikimate-5-phosphate $\rightarrow$ $\rightarrow$ prephenate $\rightarrow$ (i) phenylpyruvate $\rightarrow$ phenylalanine or (ii) $\rightarrow$ <i>p</i> -hydroxyphenylpyruvate $\rightarrow$ tyrosine. <i>See also</i> tyrosine.
18. L-Proline		Glutamic semialdehyde or α -keto- δ -aminovaleric acid			Proline $\leftrightarrow$ glutamate; proline $\leftrightarrow$ ornithine; proline $\rightarrow$ hydroxyproline. <i>See also</i> glutamic acid, ornithine.
19. L-Serine		β -Hydroxyphenylpyruvic acid			Serine $\rightarrow$ pyruvate + NH ₃ ; hydroxyphenylpyruvate $\leftrightarrow$ D-glycerate $\equiv$ (i) 2-phosphoglycerate or (ii) 3-phosphoglycerate; 3-phosphoglycerate $\leftrightarrow$ β -phosphohydroxypyruvate $\leftrightarrow$ phosphoserine $\rightarrow$ serine; phosphatidyl ethanolamine + serine $\rightarrow$ ethanolamine + phosphatidyl serine; phosphatidyl serine $\rightarrow$ CO ₂ + phosphatidyl ethanolamine; palmitaldehyde + serine $\rightarrow$ dihydrospingosine. <i>See also</i> glycine, methionine, tryptophan.
20. L-Threonine		α -Keto- β -hydroxybutyric acid			Threonine $\rightarrow$ NH ₃ + α -ketobutyrate; α -ketobutyrate $\rightarrow$ CO ₂ + propionyl CoA $\rightarrow$ methylmalonyl CoA $\rightarrow$ succinyl CoA; threonine $\rightarrow$ (i) glycine + acetaldehyde or (ii) CO ₂ + aminoacetone. <i>See also</i> aspartic acid.

TABLE 5 (continued)

Pathways of Amino Acid Metabolism			
Amino acid	Product of oxidative deamination or transamination	Product of decarboxylation	Pathways and products of metabolism
21. L-Tryptophan	β -Indolepyruvic acid	Tryptamine	Tryptophan $\rightarrow$ <i>N</i> -formylkynurenine $\rightarrow$ formate + kynurenine $\rightarrow$ 3-hydroxykynurenine $\rightarrow$ alanine + 3-hydroxyanthranilate $\rightarrow$ α -amino- β -carboxymuconate semialdehyde $\rightarrow$ CO ₂ + α -aminomuconate semialdehyde $\rightarrow$ α -aminomuconate $\rightarrow$ α -ketoadipate (see lysine for further degradation); tryptophan $\rightarrow$ 5-hydroxytryptophan $\rightarrow$ CO ₂ + 5-hydroxytryptamine (serotonin) $\rightarrow$ 5-hydroxyindoleacetic acid; indole-3-glycerol phosphate + serine $\rightarrow$ tryptophan.
22. L-Tyrosine	<i>p</i> -Hydroxyphenylpyruvic acid	Tyramine	<i>p</i> -Hydroxyphenylpyruvic acid $\rightarrow$ CO ₂ + homogentisic acid $\rightarrow$ maleylacetoacetic acid $\rightarrow$ fumarylacetoacetic acid $\rightarrow$ fumarate + acetoacetate; tyrosine $\rightarrow$ 3,4-dihydroxyphenylalanine $\rightarrow$ CO ₂ + 3,4-dihydroxyphenylethylamine $\rightarrow$ noradrenalin $\rightarrow$ adrenalin. See also phenylalanine.
23. L-Valine	α -Ketoisovaleric acid	2-Methylpropylamine	α -Ketoisovaleric acid $\rightarrow$ CO ₂ + isobutyryl CoA $\leftrightarrow$ methacrylyl CoA $\leftrightarrow$ β -hydroxybutyryl CoA $\rightarrow$ β -hydroxyisobutyrate $\leftrightarrow$ methylmalonic semialdehyde $\rightarrow$ (i) β -aminoisobutyric acid or (ii) methylmalonyl CoA (?); 2 pyruvic acid $\rightarrow$ α -acetolactate $\rightarrow$ α,β -dihydroxyisovaleric acid $\rightarrow$ α -ketoisovaleric acid $\leftrightarrow$ valine.

*Desulfination pathway for cysteine sulfenic acid $\rightarrow$ alanine + SO₂. Abbreviations: CoA = coenzyme A; ADP = adenosine diphosphate; AMP = adenosine phosphate; ATP = adenosine triphosphate; IMP = inosine phosphate; P_i = inorganic orthophosphate; PP_i = inorganic pyrophosphate.

From Altman, P. L. and Dittmer, D. S., Eds., in *Metabolism*, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1968, 426. With permission.

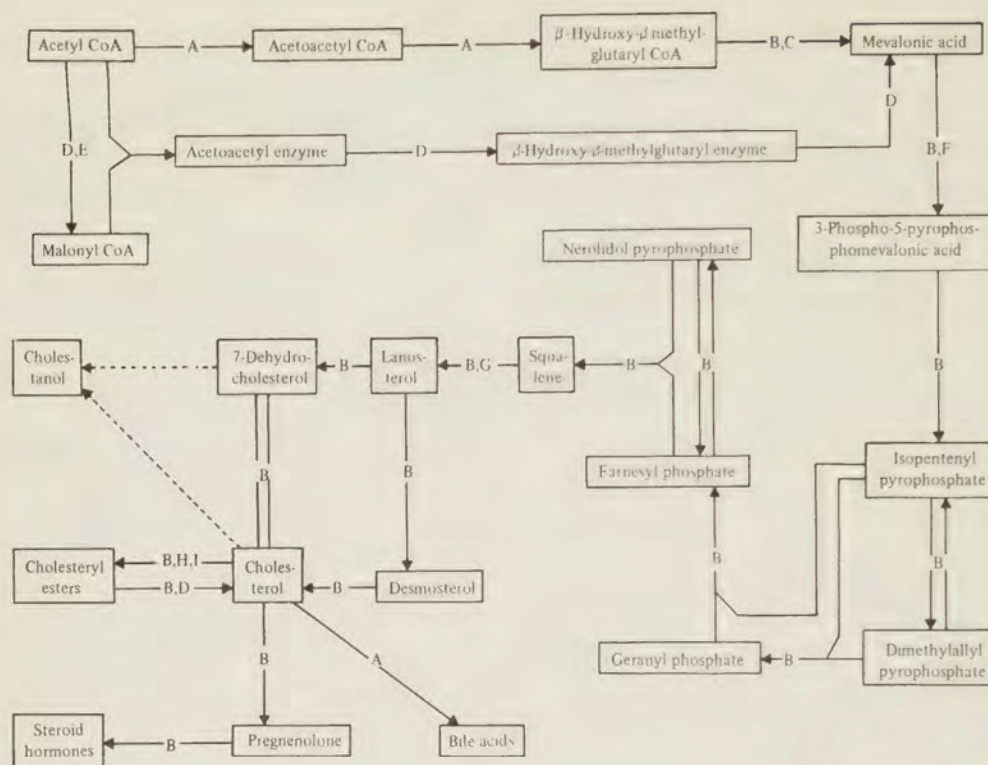


REACTIONS

- A: Occurs in mitochondria.
 B: Occurs in cytoplasm with soluble enzymes.
 C: Requires biotin, ATP, Mg^{2+} , and HCO_3^- ; citrate and isocitrate enhance carboxylation of acetyl CoA by complexing with and activating the carboxylase enzyme.
 D: β -Oxidation in mitochondria; carnitine esters are involved in transporting fatty acids into mitochondria; requires FAD and NAD^+ ; ATP or GTP may participate in preliminary extramitochondrial formation of fatty acyl CoA.
 E: Involves transacylases, ACP's β -ketoacyl-ACP synthetase, β -ketoacyl-ACP reductase, β -hydroxyacyl-ACP dehydrase, and enoyl-ACP reductase; requires NADPH (furnished mainly by pentose phosphate pathway and by oxidation of oxalacetate to pyruvate via malic acid; NADH required for the latter produced through glycolysis); yields mainly palmitic acid.
 F: Occurs in microsomes.
 G: Includes chain elongation in microsomes involving malonyl CoA and fatty acyl CoA (analogous to de novo synthesis), chain elongation in mitochondria involving acetyl CoA and fatty acyl CoA, shortening of carbon chain by 2 carbons in microsomes, and desaturation of fatty acyl CoA coupled with chain elongation to yield polyunsaturated acids (requires NADH or NADPH) in microsomes.
 H: Involves mainly pancreatic and intestinal lipases and bile salts in intestinal lumen.
 I: ω -Oxidation in intestinal lumen.

FIGURE 2. Mammalian fatty acid metabolism. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1538. With permission.)

FIGURE 3. Mammalian glycerolipid metabolism. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1539. With permission.)



REACTIONS

A: Occurs in mitochondria.

B: Occurs in microsomes.

C: Requires NADPH; not reversible; a target process in control of biosynthesis.

D: Occurs in cytoplasm with soluble enzymes.

E: Requires biotin, ATP, Mg^{2+} , and HCO_3^- ; citrate and isocitrate enhance carboxylation of acetyl CoA by complexing with and activating the carboxylase enzyme.

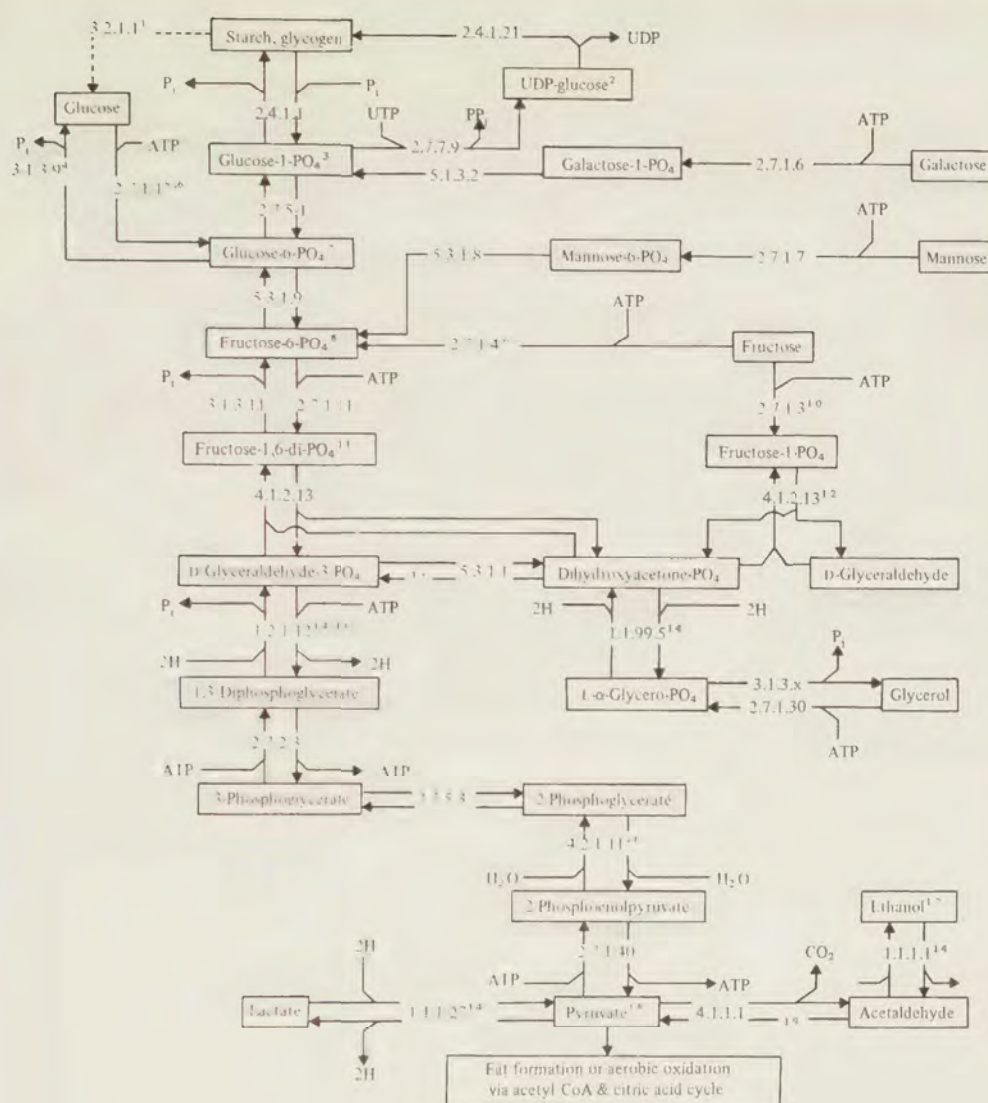
F: Requires ATP and Mn^{2+} .

G: Requires NADPH and O_2 .

H: Occurs in blood plasma.

I: Requires lecithin as the acyl donor.

FIGURE 5. Mammalian sterol metabolism. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1541. With permission.)



ENZYME KEY [COFACTOR]

- | | |
|---|---|
| 1.1.1.1 = alcohol dehydrogenase [NAD ⁺ , NADH]. | 2.7.2.3 = phosphoglycerate kinase [Mg ²⁺]. |
| 1.1.1.27 = L-lactate dehydrogenase [NAD ⁺ , NADH]. | 2.7.5.1 = phosphoglucomutase. |
| 1.1.99.5 = glycerolphosphate dehydrogenase [NAD ⁺ , NADH]. | 2.7.5.3 = phosphoglyceromutase. |
| 1.2.1.12 = glyceraldehydephosphate dehydrogenase [NAD ⁺ , NADH]. | 2.7.7.9 = glucose-1-phosphate uridylyltransferase (UDPglucose pyrophosphorylase). |
| 2.4.1.1 = α-glucan phosphorylase [AMP, Mg ²⁺ , P _i]. | 3.1.3.9 = glucose-6-phosphatase. |
| 2.4.1.21 = UDPglucose-starch glucosyltransferase. | 3.1.3.11 = hexosediphosphatase [Mg ²⁺]. |
| 2.7.1.1 = hexokinase [Mg ²⁺]. | 3.1.3.X = phosphoglycerol phosphatase. |
| 2.7.1.3 = ketohexokinase [Mg ²⁺]. | 3.2.1.1 = α-amylase. |
| 2.7.1.4 = fructokinase [Mg ²⁺]. | 4.1.1.1 = pyruvate decarboxylase [Mg ²⁺]. |
| 2.7.1.6 = galactokinase [Mg ²⁺]. | 4.1.2.13 = fructosediphosphate aldolase. |
| 2.7.1.7 = mannosokinase [Mg ²⁺]. | 4.2.1.11 = phosphopyruvate hydratase [Mg ²⁺]. |
| 2.7.1.11 = phosphofructokinase [Mg ²⁺]. | 5.1.3.2 = UDPglucose epimerase [Mg ²⁺ , UDP-glucose]. |
| 2.7.1.30 = glycerol kinase [Mg ²⁺]. | 5.3.1.1 = triosephosphate isomerase. |
| 2.7.1.40 = pyruvate kinase [K ⁺ , Mg ²⁺]. | 5.3.1.8 = mannosephosphate isomerase. |
| | 5.3.1.9 = glucosephosphate isomerase. |

FIGURE 6.

- ¹ Digestion; glycogen and/or starch are hydrolyzed to glucose in the intestinal lumen.
- ² In plants, ADP glucose is more efficiently utilized for starch synthesis than UDP glucose.
- ³ Cori ester.
- ⁴ The reaction, glycogen to glucose-6-PO₄ to blood glucose, takes place in the liver only.
- ⁵ The reaction, glucose to glucose-6-PO₄ to glycogen, takes place in liver, muscle, and other tissues.
- ⁶ Reaction assumed to be inhibited by the growth hormone plus a suprarenal cortical hormone; inhibition by these substances is blocked by insulin, thus favoring conversion of glucose to glucose-6-PO₄.
- ⁷ Robison ester.
- ⁸ Neuberg ester.
- ⁹ In all tissues.
- ¹⁰ In liver and muscle.
- ¹¹ Harden-Young ester.
- ¹² Liver aldolase also degrades fructose-1-PO₄⁻.
- ¹³ This reaction causes each step in the conversion to pyruvate to be doubled quantitatively; thus, 1 mol of glucose gives rise to 2 mol of pyruvate.
- ¹⁴ NAD⁺ acts as acceptor of released hydrogen atoms, becoming NADH in the oxidative direction of this reaction. NADH gives up hydrogen atoms and becomes NAD⁺ in the reverse direction. Hydrogen atoms accepted by NAD⁺ are passed on in turn to flavoprotein, cytochrome *c*, cytochrome oxidase, and molecular oxygen. If molecular oxygen is not sufficiently available, hydrogen atoms may be passed from NADH to pyruvate to form lactate.
- ¹⁵ Inhibited by iodoacetate.
- ¹⁶ Inhibited by fluoride.
- ¹⁷ End of fermentation in plant tissue.
- ¹⁸ Pyruvate, followed by conversion to lactate when oxygen supply is deficient (*see* Footnote 14), ends glycolysis in animal tissues. If oxygen is available, pyruvate is oxidized via the citric acid cycle.
- ¹⁹ Thiamine pyrophosphate required as cofactor in this direction only.

FIGURE 6. Carbohydrate metabolism. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1543. With permission.)

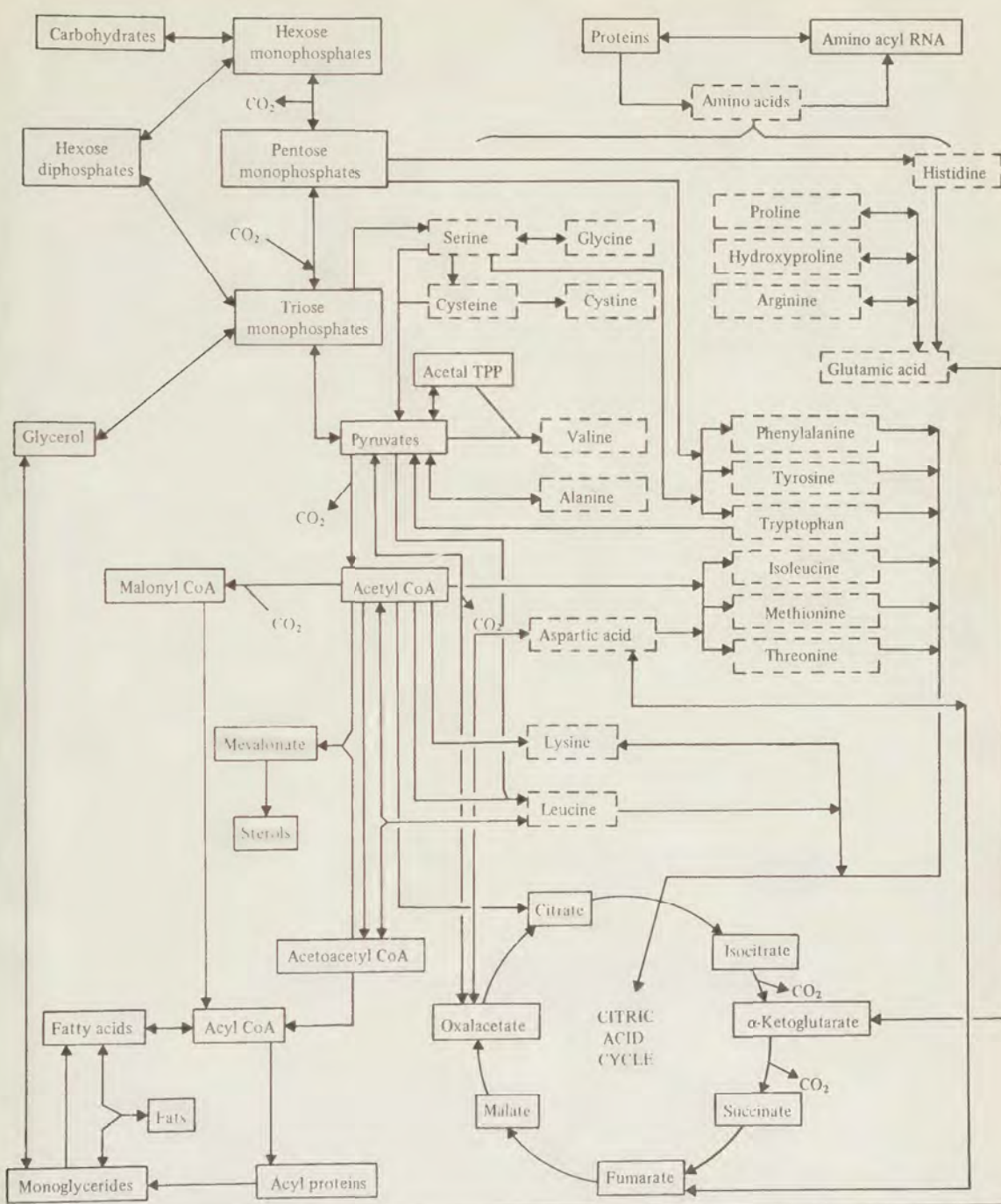


FIGURE 7. Metabolic interrelationship: Lipids, carbohydrates, and proteins. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1551. With permission.)

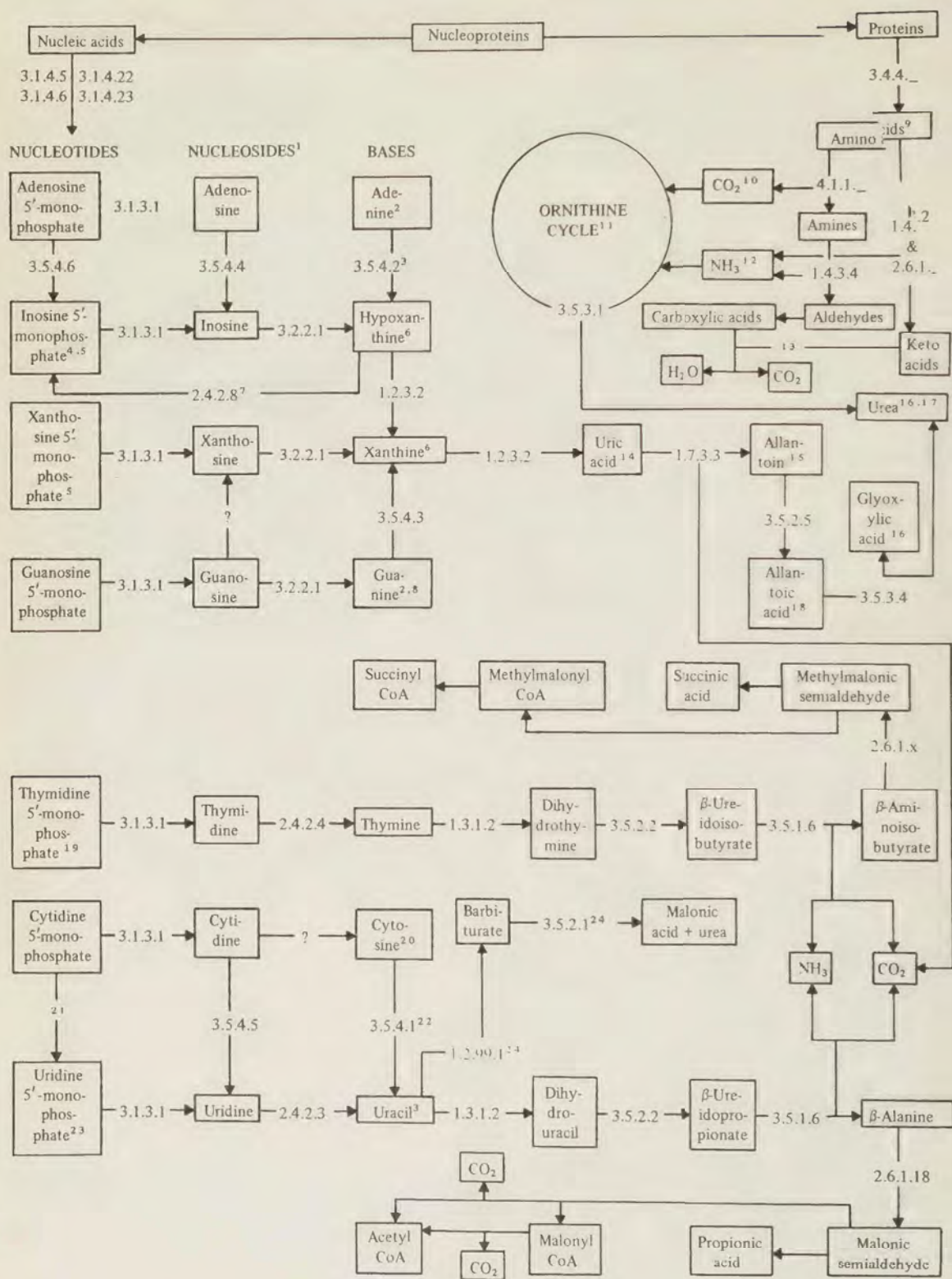


FIGURE 8.

1.2.3.2 =	xanthine oxidase.	3.1.4.23 =	ribonuclease II.
1.2.99.1 =	uracil dehydrogenase.	3.2.2.1 =	nucleosidase.
1.3.1.2 =	dihydro-uracil dehydrogenase [NADP ⁺].	3.4.4.— =	proteases.
1.4.3.2 =	L-amino-acid oxidase.	3.5.1.6 =	β -ureidopropionase.
1.4.3.4 =	monoamine oxidase.	3.5.2.1 =	barbiturase.
1.7.3.3 =	urate oxidase.	3.5.2.2 =	dihydropyrimidinase.
2.4.2.3 =	uridine phosphorylase.	3.5.2.5 =	allantoinase.
2.4.2.4 =	thymidine phosphorylase	3.5.3.1 =	arginase.
2.4.2.8 =	hypoxanthine phosphoribosyltransferase.	3.5.3.4 =	allantoicase.
2.6.1.— =	aminotransferases.	3.5.4.1 =	cytosine deaminase.
2.6.1.18 =	β -alanine aminotransferase.	3.5.4.2 =	adenine deaminase.
2.6.1.x =	aminotransferase, uncharacterized.	3.5.4.3 =	guanine deaminase.
3.1.3.1 =	alkaline phosphatase.	3.5.4.4 =	adenosine deaminase.
3.1.4.5 =	deoxyribonuclease I.	3.5.4.5 =	cytidine deaminase.
3.1.4.6 =	deoxyribonuclease II.	3.5.4.6 =	AMP deaminase.
3.1.4.22 =	ribonuclease I.	4.1.1.— =	amino acid decarboxylases.

¹ 6-Oxypurine nucleotides are split into purines and pentose phosphate by purine nucleoside phosphorylase present in tissues.

² Adenine and guanine are the major purines occurring in nucleic acids. Mammals do not require, but can synthesize, purines or pyrimidines from products of protein metabolism.

³ The route adenine $\rightarrow$ hypoxanthine is of no importance in animals; adenine deaminase [3.5.4.4] is not found to any extent in mammals.

⁴ In the biosynthesis of nucleotides, inosine 5'-monophosphate is the precursor of adenosine 5'-monophosphate and guanosine 5'-monophosphate; in uricotelic species, a portion of the inosine 5'-monophosphate is a precursor of uric acid.

⁵ Important in metabolism, but does not occur in nucleic acid.

⁶ Excreted to a limited extent by all mammals; primary end product of subjects with xanthinuria, congenital absence of xanthine oxidase [1.2.3.2]. In man, most of xanthine formed is excreted as uric acid. This is the source of $\sim 75\%$ of the urinary urate; hypoxanthine, which contributes the other 25%, is primarily a metabolic intermediate.

⁷ Recycle found in all normal mammalian tissues; absence resulting from inborn error leads to serious neurological disease.

⁸ Excreted by swine and spiders.

⁹ For more complete information on amino acid metabolism, see Table 5.

¹⁰ May enter into metabolic processes, into the ornithine cycle and be incorporated into and excreted as urea, or be excreted as CO₂.

¹¹ Urea formation in mammalian liver occurs via the ornithine cycle (Krebs-Henseleit cycle): ornithine $\rightarrow$ citrulline $\rightarrow$ arginine succinate $\rightarrow$ arginine $\rightarrow$ ornithine. CO₂ and NH₃ enter the cycle via aspartic acid at citrulline; arginine succinate is split into arginine and fumaric acid; arginine is then converted to ornithine with the release of urea.

¹² NH₃, as in the case of CO₂, is also used to synthesize many tissue constituents; hence, it may enter into many metabolic processes, be built into amino acids, be incorporated into urea and excreted, or be excreted as NH₃ across the kidney tubule.

¹³ Via the citric acid cycle (prior to entry into this cycle, sulfur-containing amino acids lose their sulfur, usually in the form of sulfate).

¹⁴ Excreted by primates, some reptiles, and some insects as the end product of purine catabolism; excreted by birds as the end product of protein, purine, and pyrimidine catabolism (no urea formation in birds).

¹⁵ Excreted by mammals other than primates, by gastropods, and by some insects.

¹⁶ Excreted by amphibians, most fishes, and freshwater lamellibranchs.

¹⁷ Excreted by mammals as the end product of amino acid metabolism.

¹⁸ Excreted by some teleosts.

¹⁹ Occurs in DNA.

²⁰ Free cytosine is excreted unchanged in animals.

²¹ Cytidine 5'-monophosphate can be converted to uridine 5'-monophosphate.

²² In yeasts and *Escherichia coli*.

²³ Occurs in RNA.

²⁴ In *Corynebacterium*, *Mycobacterium*, and U-i soil bacterium.

FIGURE 8. Pathways of nucleoprotein catabolism. (From Altman, P. L. and Dittmer, D. S., Eds., in *Biological Data Book*, Vol. III, Federation of American Societies for Experimental Biology, Bethesda, Maryland, 1973, 1552. With permission.)

Post-mortem Factors

Post-mortem factors, particularly aging, greatly influence the flavor of cooked meat. The desired flavor is not produced on cooking unaged beef, rather a metallic and astringent flavor results.^{8,4} The enzymes present in raw meat continue to act on meat components during aging and contribute towards the production of flavor precursors. This fact is supported by a number of observations:

Wood^{4,8} identified glucose, glucose-6'-phosphate, fructose-6'-phosphate, and diphosphopyridine nucleotide as reducing substances in raw meat. In an aged muscle, where autolytic processes had been allowed to continue, glucose, fructose, and ribose were the major sugars with traces of sugar phosphates. Apparently during aging, most of the sugar phosphates were enzymatically converted to sugars and inorganic phosphate.

The aging of meat principally involves the action of proteolytic enzymes on muscle proteins, which results in the tenderization of meat. It is commonly accomplished by letting beef cuts hang at temperatures of about 4°C for a period of 1 to 4 weeks. The extent of proteolysis during normal aging is slight but it can be definitely demonstrated. Aging of beef for 2 weeks results in an increase in free amino nitrogen of the nonprotein nitrogen fraction. The contribution of native tissue proteases in meat tenderization has been demonstrated by preparing rabbit and lamb muscles aseptically from freshly killed animals and preserving them under aseptic conditions at 25°C for about 150 days. After 21 days, the electrophoretic pattern of the extracted proteins showed the appearance of several new components that moved faster in the electric field. Microscopic changes were also observed, such as the longitudinal splitting of myofibrils after 50 days.^{9,5}

The effect of post-mortem myotomy on glycolysis and ultimate qualitative characteristics of porcine longissimus and rectus femoris muscles was investigated by Koch et al.^{9,6,9,7} It was observed that myotomy at or shortly after exsanguination stimulated contractile activity, significantly increased the rate of post-mortem glycolysis, and tended to decrease ultimate qualitative properties. Longissimus muscles incised at the time of exsanguination had lower pH values, glycogen, adenosine triphosphate, creatine phosphate, and higher lactate levels at all post-mortem time periods studied through 2 hr. than muscles not incised until 45 min post-mortem. The effects of myotomy were greater among normal than low quality muscles. The rates of post-mortem pH decline, levels of glycogen, lactate, glucose-6'-phosphate, adenosine triphosphate, and creatine phosphate, as influenced by the post-mortem muscle temperature and myotomy, were investigated. Muscle contraction stimulated by myotomy, particularly in the excised sample, resulted in rapid diminution of adenosine triphosphate and creatine phosphate, which in turn appeared to have contributed to the observed differences in glycolytic rate. An activating mechanism of this nature would not be expected to be as effective on a process that is already rapid as on one that is relatively slow. Thus, muscle contraction resulting from myotomy was more effective in increasing glycolytic rate of normal longissimus muscles than those of lower quality (inherent rapid glycolyzing). Also, since low temperatures (2 to 4°C) are normally used to chill pork carcasses and are known to exert an inhibitory effect upon glycolytic rate, the inhibition would be expected to have a greater effect on the faster process. Thus, the rapid heat dissipation of the rectus femoris muscles decreased glycolytic rate to a greater extent among low quality than normal muscles. Consequently, what appeared to be a discrepancy in response between the longissimus and rectus femoris muscles was in actuality a natural physiological response to differences in experimental procedure.

As has been discussed earlier, in the case of ripened sausages, proteases, lipases, and other enzymes released by micrococci and lactobacilli play a major role in the development of flavor compounds and some flavor precursors.

Nature and Concentration of Flavor Precursors Found in Meat and Their Role in the Development of Meat Flavor

During the last 2 decades, a large number of publications have dealt with the nature of flavor precursors of various types of meat. Recently, Heath¹ reported that a cold water extract of raw beef, after freeze drying and counter current dialysis, showed that only those fractions having a molecular weight of less than 200 produced any beef flavor on heating, and that fractions were composed of 32 amino acids, small

peptides, and carbohydrates such as glucose, glucosamine, fructose, and ribose. A partial list of water soluble compounds isolated from meat is given in Table 3.

The literature reports indicate that the mechanisms of meat flavor development include not only Maillard browning reactions and fatty acid oxidation but also inter- and intra-molecular cyclizations as well as numerous reactions which are made possible by the reactivity of ammonia, hydrogen sulfide, mercaptans, and other undefined intermediates, especially at elevated temperatures.^{1,7} It is generally agreed that a major portion of meat volatiles is produced as a result of interaction between sugars and amino compounds. These flavor precursors, in heated and unheated meat samples, have been extensively investigated.^{50,51,57} Quantities of individual amino compounds and sugars determined in heated and unheated lyophilized diffusate from beef, lamb, and pork are presented in Tables 6, 7, and 8. The amino compounds found in the meat of three species appear to be quite similar quantitatively as well as qualitatively, except that cysteine acid and ornithine were common only to pork and lamb, and that glutathione was present only in lamb.⁵¹ In all three cases, taurine, anserine-carnosine, and alanine were the major constituents of unheated samples, and losses during heating were relatively large. Decreases in these constituents accounted for 69, 72, and 45% of the total amount during heating of this type of compounds in beef, pork, and lamb, respectively. Other important constituents degraded during heating were glutamic acid, glycine, lysine, serine, cystine, methionine, leucine, isoleucine, and methyl histidine. The loss of essential amino acids ranged from approximately 20% in pork samples to approximately 55% in beef samples. Macy et al.⁵¹ also reported that beef contained largest amount of amino compounds, and pork the least. The percentages of the total amino compounds lost during heating were proportional to initial quantities present. Greater amounts were lost from the beef samples than from the lamb and pork samples.

Macy et al.^{50,51} reported the presence of glucose, fructose, ribose, and some unidentified carbohydrate material in beef, lamb, and pork extracts (Tables 6, 7, and 8). The concentrations of these sugars in three species were quite similar: glucose was present in greatest quantities followed by fructose, ribose, and the unknown fraction. Ribose appeared to be the most labile to heating, and fructose the most stable (Tables 6, 7, and 8).

Recently, Jarboe and Mabrouk⁶¹ carried out an extensive investigation of free amino acids, sugars, and organic acids in aqueous beef extracts. These workers confirmed the presence of glucose-6'-phosphate, fructose-6'-phosphate, fructose-1,6'-diphosphate, and adenosine-5'-monophosphate, reported earlier by Wood^{4,8} and others, and provided quantitative data on these compounds. Data of Jarboe and Mabrouk are not in agreement with the earlier work of Macy et al.,⁵¹ possibly due to differences in the state of muscle, and the analytical techniques used.

Batzer et al.⁴⁹ in 1962 identified inosinic acid, inorganic phosphate, and a glycoprotein of which the carbohydrate was glucose, as the principal components involved in the development of beef flavor on heating. The principal amino acids in the protein moiety were β -alanine and serine, with smaller quantities of glycine, glutamic acid, α -alanine, leucine and isoleucine. When mixtures of some of these amino acids were added to a solution of glucose, inosine, and inorganic phosphate, meaty flavor developed on heating in fat or water.

Comparatively, little work has been done on the contribution of water soluble components from adipose tissue in the development of meat flavor. The work done by Pepper and Pearson⁵⁸ indicates that there are some differences in proteins present in lean muscle and adipose tissue. It was suggested, therefore, that the water soluble fraction from adipose tissue may make a distinct and characteristic contribution to meat flavor above and beyond that related to the lipid portion. The authors further suggest that an additional role of adipose tissue to meat flavor could be derived from the increased number of sulfhydryl groups due to the presence of salt. Since salt is commonly used in cooking, it is possible that it may retard oxidation of these groups, thereby promoting the formation of parent sulfur compounds, which are known to play a major role in the development of cooked meat aroma. An extension of this work was recently reported by Pepper and Pearson.⁶² The dialyzable fraction from beef adipose tissue, either treated with salt or untreated and stored under either air or nitrogen, was lyophilized, fractionated by column chromatography, and analyzed by chemical procedures or thin-layer chromatographic techniques. The chromatographic profiles of dialyzable components were unaffected by salt treatments or by storage under air or nitrogen. Amino acid analysis revealed that glycine, glutamic acid, and proline comprised about 46% of the total

TABLE 6

Concentration of Various Amino Compounds and Carbohydrates in Lyophilized Diffusate from Beef before and after Heating

Compound	Concentration mg/100 g tissue	
	Unheated	Heated (boiling water for 1 hr)
Total amino compounds	161.53	71.45
Phosphoserine	0.36	0.29
Glycerophosphoethanolamine	0.02	0.01
Phosphoethanolamine	0.66	0.82
Taurine	9.05	4.02
Urea	0.01	—
Aspartic acid	0.82	0.32
Threonine	1.11	0.74
Serine + asparagine (as serine)	7.53	1.35
Glutamic acid	4.63	2.22
Glycine	2.40	1.32
Alanine	11.28	6.22
Cystine	4.37	—
Valine	2.99	1.47
Methionine	2.01	0.75
Isoleucine	2.04	0.87
Leucine	3.81	2.34
Tyrosine	1.85	0.80
Phenylalanine	1.36	0.97
NH ₃ + lysine	6.19	4.11
Histidine	4.10	4.17
Anserine + carnosine (as carnosine)	90.14	38.17
1-Methylhistidine	4.80	0.49
Total carbohydrates	48.66	28.46
Glucose	43.86	25.25
Fructose	3.56	3.21
Ribose	1.09	Trace
Unknown nucleoside (as ribose)	0.15	—

From Macy, R. L., Naumann, D. H., and Bailey, M. E., *J. Food Sci.*, 29, 142, 1964. With permission.

amino acid residues for the untreated samples as compared to 31% for the sample treated with salt. The diffusate from the salt-treated sample contained more arginine, threonine, serine, isoleucine, leucine, tyrosine, and phenylalanine (22%) than the untreated sample (9%). Only traces of sulfhydryls were evident, but positive tests were obtained for aldose and free amino groups. The diffusates from untreated adipose tissue contained about five times as many free amino groups and ten times as many aldoses as that from the salt treatment. This suggests that salt stabilized the adipose tissue from breakdown.

Thin-layer chromatography of beef adipose tissue diffusate confirmed the presence of creatine, creatinine, creatine phosphate, cytosine, uracil, and several fluorescent and phosphate-containing compounds, but purine bases, nucleosides, nucleotides, and lactic acid were absent.

Wasserman and Spinelli⁶⁰ have recently published preliminary studies on the components of adipose tissue which may be involved in the development of meat aroma (Table 9). Precursors of the characteristic beef, lamb, and pork aromas were shown to be present in the lipids extracted from adipose tissue, with a chloroform-methanol mixture. Water washing of the extract removed components involved in the formation of these odors. Amino acids and glucose were identified in the wash which produced a nonspecific roast meat aroma on heating to dryness.

TABLE 7

Concentration of Various Amino Compounds and Carbohydrates in Lyophilized Diffusate from Pork before and after Heating

Compound	Concentration mg/100 g tissue	
	Unheated	Heated (boiling water for 1 hr)
Total amino compounds	109.86	88.10
Phosphoserine	0.35	0.23
Glycerophosphoethanolamine	0.02	0.01
Phosphoethanolamine	0.45	1.89
Taurine	12.58	7.94
Aspartic acid	1.37	0.45
Threonine	0.48	0.45
Serine + asparagine (as serine)	2.95	0.53
Glutamic acid	1.95	1.19
Proline	0.64	0.24
Glycine	2.75	1.58
Alanine	4.19	2.80
Cystine	2.11	0.89
Valine	0.30	0.20
Methionine	0.69	1.00
Isoleucine	1.03	0.83
Leucine	1.68	1.00
Tyrosine	0.56	0.37
Phenylalanine	0.51	0.39
Ornithine	Trace	0.56
NH ₃ + lysine	4.27	4.06
Histidine	2.55	3.11
Anserine + carnosine (as carnosine)	67.94	58.38
1-Methylhistidine	0.49	Trace
Total carbohydrates	45.90	30.04
Glucose	43.56	28.02
Fructose	2.08	2.02
Ribose	0.20	Trace
Unknown nucleoside (as ribose)	0.06	—

From Macy, R. L., Naumann, D. H., and Bailey, M. E., *J. Food Sci.*, 29, 142, 1964. With permission.

Since adipose tissue is composed of metabolically active cells, it is not surprising that a number of amino acids and at least one sugar component have been identified in tissue extracts. The absence of purine bases, nucleosides, and nucleotides in beef adipose tissue diffusate, reported by Pepper and Pearson,⁶² may be due to inadequate detection techniques, as nucleic acids are essential cellular constituents and their metabolic and breakdown products are usually found in living cells.

As reported earlier, Batzer et al.⁴⁹ found that inosinic acid, along with glucose and unidentified glycoprotein, was necessary for the development of meaty flavor in beef. Inosinic acid is produced from adenosine 5'-triphosphate post-mortem. Nucleotides are also potential precursors of free ribose and ribose phosphate, which have been implicated in Maillard browning reactions. Macy et al.⁵⁶ investigated the effect of cooking on nucleotides, nucleosides, and bases of beef, pork, and lamb. The effect of cooking on beef and pork nucleotides is given in Table 10. These workers reported that inosinic acid was the prominent nucleotide in all three species and it was degraded by heating. Adenylic acid increased during cooking in meat from all three species, possibly due to the hydrolysis of adenosine di- and triphosphate and nucleic

TABLE 8

Concentration of Various Amino Compounds and Carbohydrates in Lyophilized Diffusate from Lamb before and after Heating

	Concentration mg/100 g tissue	
	Unheated	Heated (boiling water for 1 hr)
Total amino compounds	130.01	81.46
Phosphoserine	0.30	0.22
Glycerophosphoethanolamine	0.01	—
Phosphoethanolamine	1.15	1.90
Taurine	26.25	16.47
Urea	Trace	—
Aspartic acid	1.52	1.36
Threonine	3.14	1.36
Serine + asparagine (as serine)	4.74	2.26
Glutamic acid	6.08	2.85
Proline	3.05	0.83
Glycine	4.30	2.25
Alanine	9.18	6.53
Cystine	6.46	5.24
Valine	0.44	0.18
Methionine	2.37	1.53
Isoleucine	2.79	1.87
Leucine	5.62	3.06
Tyrosine	1.98	1.67
Phenylalanine	1.85	1.12
Ornithine	0.95	0.65
NH ₃ + lysine	10.51	5.59
Histidine	9.70	7.16
Anserine + carnosine (as carnosine)	25.55	16.28
1-Methylhistidine	2.07	1.08
γ -Amino- <i>n</i> -butyric acid	Trace	—
Total carbohydrates	36.42	23.94
Glucose	32.87	21.33
Fructose	2.68	2.61
Ribose	0.52	Trace
Unknown nucleoside (as ribose)	0.35	—

From Macy, R. L., Naumann, D. H., and Bailey, M. E., *J. Food Sci.*, 29, 142, 1964. With permission.

acids. Cytidylic, uridylic, and guanylic acids were present in relatively low concentrations in meat from all three species and changed little during cooking.

It is well known that stiffening of muscle post-mortem is related to decrease in adenosine-5'-triphosphate (ATP) content.⁹⁸⁻¹⁰³ The formation of adenosine-5'-monophosphate (AMP) results from the removal of two terminal phosphate groups of ATP by the combined catalytic action of ATPase and myokinase. AMP is rapidly deaminated to inosinic acid (IMP) by the catalytic action of AMP deaminase. Further degradation of IMP into inosine and hypoxanthine occurs during prolonged storage. The details of these metabolic reactions may be found in Figure 8.

Howard et al.¹⁰⁴ studied nucleotide breakdown in beef tissue and found that with progressive aging the hypoxanthine levels rose to 1.5 to 2.0 $\mu\text{mol/g}$ of meat. Lee and Webster¹⁰⁵ studied the effect of temperature, freezing, frozen storage, thawing, and pH on hypoxanthine production in beef tissue and

TABLE 9

Amino Acid Composition of the Water-soluble Fraction
from Chloroform:Methanol Extracts of Adipose Tissue^{6,9}

Compound	Concentration $\mu\text{mol}/100\text{ g tissue}$		
	Pork	Beef	Lamb
Cysteic acid	2.35	1.95	1.50
Taurine	19.40	9.20	34.70
Urea	44.60	39.15	—
Aspartic acid	0.85	3.60	3.25
Asparagine + threonine	10.00	3.85	8.85
Serine	7.00	3.90	8.40
Glutamic acid	4.50	4.90	8.25
Proline	7.05	4.45	10.35
Glycine	16.85	13.20	35.25
Alanine	39.55	25.75	45.40
Valine	5.80	3.90	8.65
Methionine	1.65	0.90	2.30
Isoleucine	5.45	6.55	11.40
Leucine	8.10	4.35	11.30
Tyrosine	4.20	1.65	4.45
Phenylalanine	2.95	2.50	5.80
Ammonia	3.65	2.90	1.15
Lysine	0.75	0.95	1.90
Anserine	0.80	0.50	1.55
Carnosine	5.90	0.85	0.75
Arginine	1.70	0.80	0.11

TABLE 10

Ratios of Sums of Individual Nucleotides and Total Nucleotides of Cooked
Relative to those of Raw Beef and Pork^{5,5}

Constituent	Ratio of chemical constituents in cooked and raw meat (raw meat = 1.0)			
	Beef		Pork	
	49°C	71°C	49°C	71°C
Cytidylic acid	0.81	0.99	0.67	0.58
Adenylic acid	1.46	3.38	0.80	1.81
Uridylic acid	0.59	0.75	0.83	0.84
Inosinic acid	0.75	0.61	0.90	0.78
Guanylic acid	0.55	1.41	0.75	0.67
Sum of individual nucleotides	0.77	0.74	0.87	0.83

TABLE 11

Post-mortem Changes in the Concentrations of Nucleotides and Related Compounds in Porcine Longissimus Muscle^{5,9}

Breeds	Post-mortem time (hr)	Concentration $\mu\text{mol/g}$ wet tissue					
		Inosine + hypoxanthine	DPN ⁺ + DPNH	5'-IMP	5'-IDP	5'-ADP	5'-ATP
Poland China (n=2)	0	—	0.62	2.11	—	1.09	4.13
	3	1.65	0.10	5.48	0.59	0.38	0.16
	24	2.12	Trace	5.17	0.54	0.63	Trace
	72	2.75	Trace	5.03	0.69	0.54	Trace
	144	2.73	Trace	4.47	0.46	0.36	Trace
Chester White (n=2)	0	—	0.66	0.78	—	0.90	6.04
	3	1.26	0.49	5.67	0.42	0.33	0.38
	24	1.72	0.30	5.44	0.69	0.31	0.21
	72	2.07	0.17	5.20	0.79	0.37	0.23
	144	2.90	0.15	4.59	0.57	0.42	0.11

reported that an increase in temperature and a higher ultimate pH or thawing of frozen samples stimulated the rate of hypoxanthine production.

Tsai et al.^{5,9} investigated the post-mortem metabolism of nucleotides in porcine longissimus muscle. The post-mortem changes in the concentrations of nucleotides and related compounds post-mortem are reported in Table 11. Tsai and co-workers observed that AMP deaminase activity decreased considerably during storage, whereas adenosine deaminase activity remained nearly the same during storage. On the other hand, 5'-nucleosidase activity seemed to increase during the first few hours of storage and then remained constant for several days before declining. The direct consequence of decrease in AMP deaminase activity during storage resulted in the stabilization of IMP concentration within the first few hours of post-mortem. On the other hand, inosine + hypoxanthine concentration in muscle continued to increase for several days, as would be expected from nearly constant adenosine deaminase activity during storage (Table 11).

During the ripening of dry sausage, the concentration of water soluble nitrogenous compounds increases and can reach values up to 25% of the total nitrogen.^{10,6-10,8} The nature and concentration of these compounds such as amino acids, peptides, nucleotides, and nucleosides determine, to a large extent, the final dry sausage flavor.^{10,9}

Pohja and Niinivaara^{11,0} and Sajber et al.^{11,1} reported that free amino acid production in dry sausage is at least partly due to bacterial protease activity. Although native muscle proteases may continue to hydrolyze proteins in dry sausage, if not inactivated during processing, their contribution in the liberation of free amino acids would be comparatively smaller to bacterial proteases. Cantoni et al.^{11,2} investigated the fate of inosinic acid during dry sausage ripening and reported that inosinic acid was converted to inosine nucleoside and hypoxanthine, respectively, by the action of phosphomonoesterase and nucleosidase. Dierick et al.^{11,3} investigated changes in nonprotein nitrogen compounds during dry sausage ripening. Two batches of sausages were prepared: Batch 1 was a normal sausage preparation;^{11,4} in batch 2 a starter culture, containing lactobacilli and micrococci, and 1% less sugar were added. The concentration of nonprotein nitrogen compounds (Table 12) and free amino acids (Table 13) in the two batches were monitored during ripening. These data show that during the first 3 days of ripening, the rate of free amino acid production is maximal and exceeds the rate of ammonia production and peptide production from proteins. During this period intensive carbohydrate metabolism and bacterial growth also takes place.^{11,4} Subsequent to this, the rate of ammonia production increases but remains lower than amino acid production, whereas the concentration of peptides decreases. The concentration changes for individual amino acids during ripening (Table 13) show that the major amino acids responsible for the increase in total α -amino nitrogen were alanine, leucine, threonine, valine, serine, glycine, and proline. It was also reported

TABLE 12

Concentration of NPN Compounds at Various Stages of Dry Sausage Ripening (mg N/100 g dry matter)

Compound	Batch 1 Stage of ripening (days)				Batch 2 Stage of ripening (days)			
	0-3	3-15	15-36	0-36	0-3	3-15	15-36	0-36
Ammonia	6	28	18	52	2	34	12	48
Free amino acids	47	46	21	114	45	30	72	147
Peptide-nitrogen	34	-43	-7	-16	10	-67	-55	-112
Nucleotide-nitrogen	-1	-20	-1	-22	-16	-5	-2	-23
Nucleoside-nitrogen	8	37	5	50	11	33	14	58

From Dierick, N., Vandekerckhove, P., and Demeyer, D., *J. Food Sci.*, 39, 301, 1974. With permission.

TABLE 13

Concentration of Free Amino Acids at Three Stages of Ripening (mg α -NH₂-N/100 g dry matter)

Free amino acid	Batch 1 Stage of ripening (days)			Batch 2 Stage of ripening (days)		
	0	15	36	0	15	36
Aspartic acid	0.74	1.90	5.30	0	3.79	7.25
Threonine	0.82	3.30	25.20	0	4.95	6.94
Serine	1.73	5.50	9.10	0	7.20	9.85
Glutamic acid	19.00	7.20	5.60	25.40	24.30	18.30
Proline	0	3.40	5.50	0	5.72	6.45
Glycine	3.00	6.15	8.80	0	7.65	14.20
Alanine	10.20	20.90	25.20	3.92	23.10	22.30
Valine	1.44	6.35	8.85	0	7.41	11.95
Methionine	0.56	2.72	3.84	1.60	3.38	5.26
Isoleucine	1.60	3.74	5.45	2.24	4.50	8.10
Leucine	1.06	11.50	13.30	8.30	13.20	17.60
Phenylalanine	0.93	4.50	5.25	2.98	5.00	7.15
Lysine	2.07	4.67	6.35	2.30	3.94	6.46
Histidine	0.73	1.46	0.01	2.77	2.20	0
Tyrosine	0.77	0	0	0	0	0.30
γ -Amino butyric acid	0	2.72	4.07	1.22	7.89	12.50
Ornithine	1.16	0	0	1.98	0	0.83

From Dierick, N., Vandekerckhove, P., and Demeyer, D., *J. Food Sci.*, 39, 301, 1974. With permission.

that during sausage ripening added glutamate disappeared, and at least partly decarboxylated to γ -amino butyric acid. Langner,¹¹⁵ based on the observation that glutamate is metabolized during dry sausage ripening, questioned its incorporation as a flavor additive in dry sausage. Other amino acids may be decarboxylated during dry sausage ripening, as indicated by the disappearance of histidine, tyrosine, and ornithine. The decarboxylation products of these amino acids are histamine, tyramine, and putrescine, respectively. Data reported by Langner¹¹⁶ and Reuter and Langner¹¹⁷ are in contrast with those of Dierick et al.¹¹³ These workers reported an increase in these amino acids. In order to prove the validity of their observation that histidine, tyrosine, and ornithine are indeed decarboxylated during sausage ripening, Dierick et al.¹¹³ determined the concentration of highly basic amines in the ripened samples. Although very small amounts were detected, the concentration of histamine, tyramine, and putrescine was increased at least tenfold, the rate of increase being maximal during the first three days of ripening. Cadaverine, a decarboxylated product of lysine, was also detected in significant quantities. The presence of starter culture in batch 2 produced no striking differences in the nature and concentration of nonprotein nitrogen compounds during ripening, except for a higher final concentration of free amino acids, coupled to a lower peptide-nitrogen and lower increase in threonine concentration (Tables 12 and 13). The lower concentration of amino acids in batch 2 may suggest a higher exopeptidase activity in batch 2.

MECHANISMS OF MEAT FLAVOR DEVELOPMENT

During the heat processing of meat, the types of reactions which appear to be responsible for the development of flavor compounds include:

1. Autoxidation, hydrolysis, dehydration, and decarboxylation of fats giving rise to aldehydes, fatty acids, lactones, and ketones.
2. The nonfatty components may undergo glycoside splitting, oxidation and decomposition to yield volatile and nonvolatile compounds.
3. The complex degradation products of sugars and sugar-amine interactions.

These chemical reactions are discussed in detail in this section. The reactions are divided into subheadings: amino acids, carbohydrates, and lipids, depending on which is the substrate of the reaction.

Amino Acids

The thermal decomposition of amino acids has not been studied at temperatures normally attained in cooking of meat, but several investigations have been made at temperatures of 200°C and above. Based on the energies of activation of these reactions, it is conceivable that some of these reactions would take place at lower (cooking) temperatures.

The thermal decomposition products of alanine, β -alanine, valine, leucine, and isoleucine, which were heated at or above 220°C, were identified by Lien and Nawar (Table 14).^{118,119} They have also proposed mechanisms of their formation (Figure 9).

Olefins are produced by the decarboxylation and deamination of aliphatic amino acids; the corresponding paraffins result upon reaction with hydrogen. By this process isobutylene and isobutane are produced from valine, 3-methyl-1-butene and isopentane from leucine, and 2-methyl-1-butene from isoleucine. In addition, the formation of propane and propene from valine, isobutane and isobutylene from leucine, and butane and butene from isoleucine, may be explained by cleavage of carbon-carbon bonds beta to carbonyl group of the amino acid to form a free radical which can then accept or lose a hydrogen atom.

Primary amines are produced by decarboxylation, while the corresponding aldehydes result from both deamination and decarbonylation of the amino acid. The formation of isobutyraldehyde and isobutylamine from leucine cannot be explained by the deamination and decarbonylation reactions.¹¹⁹ The imines formed from the heat degradation of amino acids probably originate from the reaction of carbonyl compounds with primary amines. Aliphatic ketones react with amines more slowly than aldehydes to form imines which necessitates the use of higher reaction temperatures and longer reaction times than required for the aldehyde.¹²⁰ This is probably the reason why Lien and Nawar^{118,119} found only aldimines and

TABLE 14

Thermal Decomposition Products from Alanine, β -Alanine, Valine, Leucine, and Isoleucine^{118,119}

Alanine	β -Alanine	Valine	Leucine	Isoleucine
Ammonia	Ammonia	Ammonia	Ammonia	Ammonia
Carbon dioxide	Carbon dioxide	Carbon dioxide	Carbon dioxide	Carbon dioxide
Carbon monoxide	Carbon monoxide	Carbon monoxide	Carbon monoxide	Carbon monoxide
Ethane	Ethane	Propane	Isobutane	Butane
Ethene	Propene	Isobutene	Isobutylene	Butene
Propene	Acetonitrile	Isobutylene	Isopentane	Isopentane
2-Butene	Acetone	Isobutylene	3-Methyl-1-butene	2-Methyl-1-butene
Acetaldehyde	Pyridine	Acetone	Acetone	2-Butanone
Ethylamine	3-Picoline	Isobutyraldehyde	Isobutyraldehyde	2-Methylbutyraldehyde
N-Ethylidene-ethylamine	2,4-Lutidine	Isobutylamine	Isovaleraldehyde	2-Methylbutylamine
2-Methyl-5-ethylpyridine	3,5-Lutidine	N-Isobutylideneisobutylamine	Isobutylamine	N-(2-methylbutylidene)-2-methylbutylamine
N-Ethylpropionamide	2,3,5-Trimethylpyridine	Diisobutylamine	Isoamylamine	bis(2-Methylbutyl)amine
Propionamide	Poly- β -alanine		N-Isobutylidene-isoamylamine	
			N-Isoamylidene-isoamylamine	
			Diisocamylamine	

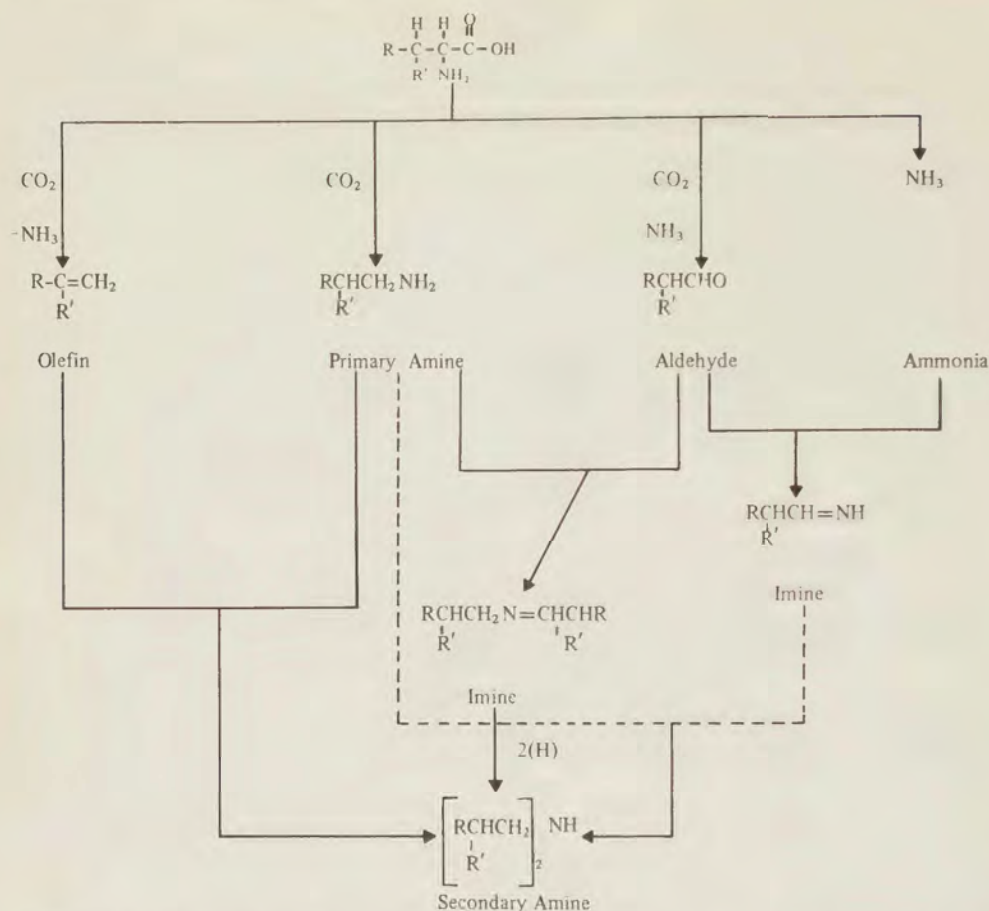


FIGURE 9. Suggested mechanisms for the production of heat degradation products from neutral aliphatic amino acids. (see Table 14) (From Lien, Y. C. and Nawar, W. W., *J. Food Sci.*, 39, 911, 1974. With permission.)

not ketimines in their investigation on thermal decomposition products of aliphatic amino acids. Thus the reaction of isobutyraldehyde and isobutylamine, both thermal decomposition products of valine, resulted in the formation of *N*-isobutylidene-isobutylamine. Similarly, *N*-isobutylidene-isoamylamine and *N*-(2-methylbutylidene)-2-methylbutylamine were produced from thermal decomposition of leucine and isoleucine, respectively. Aliphatic aldehydes can also react with ammonia to first form simple addition compounds called "aldehyde ammonias." These compounds are unstable and easily decompose to the original constituents, or lose water to form imines.¹²¹ The secondary amines diisobutylamine, diisoamylamine, and bis(2-methylbutyl)-amine result from the addition of hydrogen to the above mentioned imines. An alternative pathway, however, may be the reaction of primary amines with imines to additional products which can be reduced to secondary amines.¹²² A third possibility may result from the addition of primary amines to olefins.¹²³ A characteristic imine, *N*-ethylidene-ethylamine, was detected in the heat breakdown products of alanine. This imine probably resulted from the reaction of acetaldehyde and ethylamine;¹¹⁹ however, the secondary amine, which was present in the degradation products of valine and leucine, was absent in alanine (Table 14).

The formation of 2-methyl-5-ethylpyridine in the case of alanine may arise from the aldol condensation of acetaldehyde to form crotonaldehyde, followed by dimerization in the presence of ammonia. The most probable reaction mechanism of pyridine synthesis is via Tschitschibabin reactions.¹²¹ This mechanism

involves the reaction of aldehyde with ammonia to form imine, followed by aldol-type condensation to produce pyridine derivatives. The absence of pyridine derivatives in pyrolyzed valine, leucine, or isoleucine is probably due to their branched chain structure, which may make the aldol-type condensation difficult.¹²⁰ The two amides, propionamide and *N*-ethylpropionamide, also not found in the case of valine, leucine, and isoleucine, were produced in relatively large quantities from alanine. The formation of these two compounds may result from the breakdown of 3,6-dimethyl-2,5-diketopiperazine, which is the dehydration product from two molecules of alanine.

The β -amino acids decompose at lower temperatures than α -amino acids. β -Alanine for example loses its crystalline appearance and develops a yellow color at 200°C, whereas α -amino acids develop yellow color at about 250°C. The decomposition products of β -alanine were also significantly different from those of α -amino acids (Table 14).

Merritt and Robertson¹²⁴ listed the major compounds formed on pyrolyzing various common amino acids. Among these compounds benzene, toluene, and ethylbenzene were obtained on pyrolyzing phenylalanine and the corresponding hydroxy-compounds were derived from tyrosine. Imidazole derivatives were formed from histidine.¹²⁵ A variety of nitriles have also been reported.¹²⁶ Most interesting, however, is Merritt's observation that the compounds formed on pyrolyzing a peptide depended on the sequence of amino acids. Although only dipeptides were used in this study, this presumably holds true for longer peptides and may explain the formation of flavor compounds not obtained on heating model systems of single amino acids.¹²⁷

Carbohydrates

The heat degradation products of carbohydrates, particularly sugars, are important flavoring ingredients, and have been studied extensively. On heating, sugars lose their bound water between 100 to 130°C, without alteration in molecular structure. With increasing temperature, 150 to 180°C, a molecule of water splits from sugars forming anhydrides, and further heating, 190 to 220°C, results in the removal of second water molecule with the formation of furfural from pentoses and hydroxymethylfurfural from hexoses.

Table 15 lists some compounds that have been reported as carbohydrate caramelization and pyrolysis products.¹²⁸ Following are likely reaction steps in sugar caramelization: enolization of aldoses with the production of more reactive 2-ketoses; dehydration of ketohexoses, without fission, to 5-(hydroxymethyl)-2-furaldehyde, and dehydration of pentoses similarly to 2-furaldehyde; hydrolytic fission of furaldehydes or intermediates leading to furaldehydes to yield, for example, formic acid and levulinic acid from hexoses, fission of 2-ketoses to yield dihydroxyacetone and glyceraldehyde, glycolaldehyde and four-carbon carbonyl compounds; dehydration of the trioses to yield acetol and pyruvaldehyde; dismutation of biose, trioses, tetroses, and their dehydration products to yield lactic aldehyde, pyruvic aldehyde, lactic acid, glycolic acid, acetaldehyde, acetic acid, formaldehyde, formic acid, acetoin, and diacetyl; self- and cross-condensations of aldehydes and ketones containing active hydrogen; reversion of aldoses and ketoses to di-, tri-, and higher oligosaccharides; dimerization of fructose to fructose anhydrides; cyclodehydration of aldoses to glycosans, and then polymerization; and enolization and dehydration of the synthetic oligosaccharides. Some of these reactions are referred to again under amino acid-carbohydrate reactions.

Amino Acid-Carbohydrate Reactions

Although amino acids and carbohydrates degrade on heating to form flavor compounds, in meat they exist in close proximity and interact with each other to form a variety of compounds.¹²⁷ Flavor production by nonenzymatic browning in foods proceeds mainly from reactions of reducing sugars with amines, amino acids, peptides, and proteins.

Aldoses react with primary or secondary amines to give aldosylamines. Under the influence of a suitable catalyst, aldosylamines rearrange to form ketoseamines. This reaction is known as the Amadori rearrangement. Ketoseamines can also be formed directly from aldoses and amines. Ketoseamines are distinguished from aldosylamines by their relatively strong reducing properties and by the formation of sugar degradation products, instead of parent hexoses, on acid hydrolysis. The chemical reactions leading to the formation of flavor compounds, i.e., furaldehydes, acetaldehyde, pyruvic aldehyde, diacetyl, acetic acid, and others, are outlined in Figure 10. These compounds are also produced as caramelization products

TABLE 15

Carbohydrate Caramelization and Dehydration Products^{1,2,8}

Carbon monoxide	Acetoin
Carbon dioxide	Diacetyl
Formaldehyde	Hydroxydiacetyl
Formic acid	C-Methyl triose-reductone
Acetaldehyde	Levulinic acid
Acetic acid	Furan
Glycolaldehyde	2-Methyl furan
Glyoxal	2,5-dimethyl furan
Glyoxylic acid	Furfuryl alcohol
Lactic aldehyde	2-Furoic acid
Lactic acid	2-Furaldehyde
Acrolein	5-Methyl-2-furaldehyde
Acrylic acid	5-(Hydroxymethyl-2-furaldehyde
Pyruvaldehyde	2-Furyl methyl ketone
Pyruvic acid	2-Furyl hydroxymethyl ketone
Acetone	Isomaltol
Acetol	4-Hydroxy-2,5-dimethyl-3-(2H)-furanone
Dihydroxyacetone	Reductic acid
Glyceraldehyde	1-Methylcyclopentenol(2)-one(3)
Triose-reductone	Maltol
Mesoxalic dialdehyde	

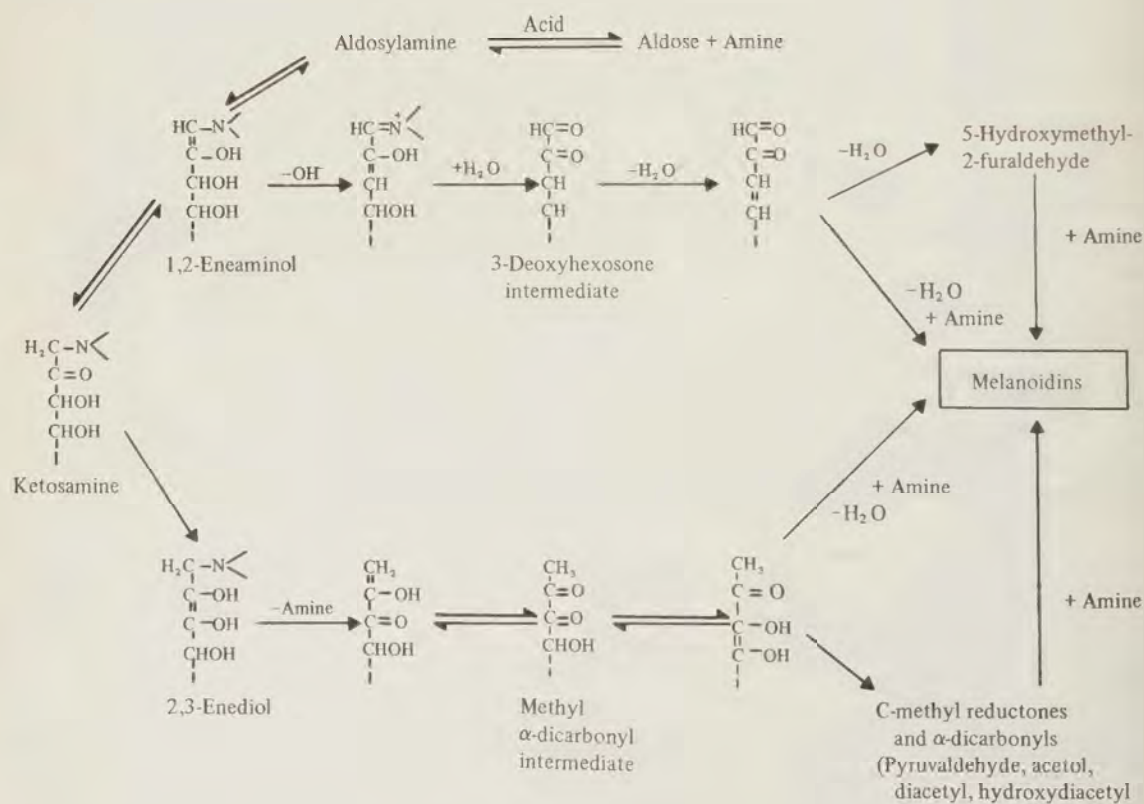


FIGURE 10. Mechanism of the Maillard reactions. (From Hodge, J. E., in *The Chemistry and Physiology of Flavors*, Schultz, H. W., Day, E. A., and Libbey, L. M., Eds., Avi Publishing, Westport, Conn., 1967, 465. With permission.)

of sugars. In the absence of amines it may be assumed that 1,2-enolization of sugars leads to the same transformations shown in Figure 10.¹²⁸ The hydroxyl group on C-3 and C-1 would be eliminated (at higher temperatures or under more acidic or alkaline reaction conditions) in the same way shown for glycosylamines and ketoseamines. For example, maltol is formed from maltose by heat alone,¹³¹ and also in the presence of amino compounds,^{129,132} but the condensation with amino compounds allows the enolizations and eliminations to take place near neutral pH and at much lower temperature.

The Maillard browning reactions have been reviewed by Reynolds^{129,130} and Hodge.¹²⁹ Both aldosylamine and ketoseamine reaction schemes provide active compounds for the degradation of α -amino acids to aldehydes and ketones of one less carbon atom by the Strecker degradation mechanism. In this reaction it is required that the amino group must be α to the carboxyl group and that the carbonyl compound must contain a $-C:O-C:O$ or a $-C:O-(CH:CH)_n-C:O$ grouping. The reaction is illustrated by the following equation:



If the hydrogen on the α -carbon of the amino acid is substituted, a ketone is produced. For example, acetone is formed from α -aminoisobutyric acid. 2-Furaldehyde produced in Maillard browning reactions participates in the Strecker degradation reaction because it yields a α -diketone and a conjugated dicarbonyl compound by hydrolytic ring opening. Several α -dicarbonyl compounds that are produced by simple heat treatment of carbohydrates are listed in Table 15. Each of these may react amino compounds to produce aldehydes. Furthermore, sugar derived, nonvolatile α -dicarbonyl compounds (Figure 10), and dehydroascorbic acid are likely compounds for inducing the Strecker degradation. The major Strecker degradation aldehydes produced from common amino acids include: formaldehyde, acetaldehyde, propionaldehyde, isobutyraldehyde, isovaleraldehyde, 2-methyl butanal, glycolic aldehyde, lactic aldehyde, methional, benzaldehyde, and several others.¹²⁸

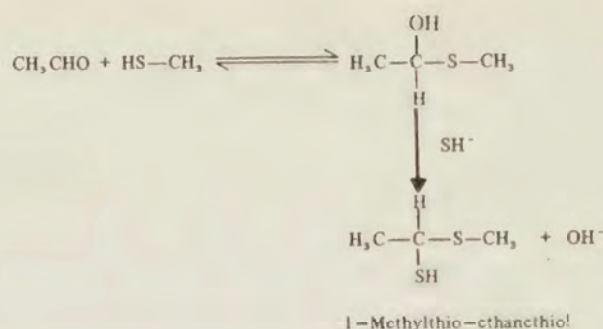
The origins of other important meat volatiles include:

Maillard reaction products of sulfur containing amino acids and sugars. Arroyo and Lillard¹³³ identified a number of sulfur compounds from methionine, cysteine, and cystine-glucose mixtures including methyl mercaptan, ethyl mercaptan, propyl mercaptan, dimethyl sulfide, dimethyl disulfide, and pentyl mercaptan.

Thialdine has been reported to form from ammonia, hydrogen sulfide, and acetaldehyde.²⁹ These three compounds have been reported in the headspace of beef broth. Brinkman et al.²⁹ believe that in view of the ease of thialdine formation from the above mentioned compounds, it is almost certainly present in the broth or the headspace and can be considered as a contributor to beef broth flavor.

Another interesting sulfur compound first identified by Herz³ in cooked beef is 2,5-dimethyl-1,3,4-trithiolane. This compound has a sulfide odor in concentrated form, but when diluted gives a meaty aroma. It may well be an important contributor to the aroma of boiled beef. Asinger and Thiele^{134,135} have described the synthesis of this compound from acetone, hydrogen sulfide, and sulfur in diisobutylamine solvent. Similar conditions could exist locally during boiling of beef, where evolution of both hydrogen sulfide and ammonia occurs. Herz³ hypothesizes that the sulfur atom could be supplied by active methionine (adenosylmethionine), and acetone could arise from many compounds in beef.

The findings about the origin of thialdine and trithiolane prompted an investigation into the origin of 1-methylthio-ethanethiol. This compound has particularly interesting organoleptic properties; in very dilute aqueous solutions (1 to 5 $\mu\text{g/l}$) it has a meaty rather than its usual onion-like odor.²⁹ Scutte and Koenders¹³⁶ have proposed a reaction mechanism for the synthesis of this compound in beef broth from known beef components. The proposed system has been substantiated experimentally. 1-Methylthio-ethanethiol is formed when acetaldehyde, methanethiol, and hydrogen sulfide are heated in aqueous solution at pH 6. These immediate precursors are in turn generated under the same conditions from alanine, methionine and cysteine in the presence of Strecker degradation agent such as pyruvaldehyde. The proposed mechanism may be represented as follows:



Another important class of compounds identified in cooked meat aroma are pyrazines. Pyrazines are known to arise from the condensation of sugars and amino acids. The necessary precursors, that is, amino acids and sugars, are available in uncooked meat and pyrazine formation should occur readily upon heating. Koehler and Odell¹³⁸ have recently investigated the factors affecting the formation of pyrazine compounds in sugar-amine interaction.

Lipids

The role of lipids in the development of the characteristic aroma of meat is a subject of some controversy. The ability of fat to dissolve and retain aroma that originates outside the fat is well known and demonstrated in the case of smoked meat products. Fat trimmed from uncooked meat has little characteristic flavor. It has been reported that rendering of fat trimmed from uncooked meat at low temperature does not produce species specific flavor. However, if rendered at normal cooking temperatures species flavor develops with little or no basic meat character.⁹⁸ In the work carried out at Kohnstamm and Co.,⁹⁸ rendered fat was steam distilled and distillate solvent extract. Gas-liquid chromatographic studies of meat fat volatiles showed that the characteristic flavor of cooked meat fat depended largely on unsaturated aldehydes. There was considerable qualitative similarity between meat fat volatiles of different species but the quantitative picture showed considerable differences. Fatty acids, methyl ketones and saturated aldehydes were also found and are believed to play a supporting role.⁹⁸ Wasserman and Spinelli¹³⁸ investigated the role of adipose tissue constituents in the development of species specific flavor. Adipose tissue, which yields the characteristic aroma of the species on heating, was extracted with chloroform-methanol (2:1), and the extract was washed with water. The original extract retained the species specific aroma but in the water-washed residue the characteristic aroma was either missing or greatly reduced. The water soluble material extracted from adipose tissue contained amino acids and at least glucose in the carbohydrate fraction. A typical lean meat aroma developed on heating this fraction.

Lipids may contribute to meat flavor in several ways:

Lipids may act as solvent, trapping aroma components produced elsewhere. Thus, the aroma of water-washed pork lipid extract still had a "piggy" note when heat was first applied.¹³⁸ This may be due to 5 α -androst-16-en-3-one, the boar odor identified by Patterson,⁸⁵ which is dissolved in the lipids of the animal.

Lipid components or degradation products may react with compounds produced in the lean meat.

Lien and Nawar¹³⁹ investigated the interaction of amino acids and triglycerides, using valine and tricaproin as a simple model system, under nonoxidative conditions. The decomposition products identified from valine-tricaproin mixtures heated at 270°C for 1 hr are listed in Table 16. A scheme for the interaction of valine and tricaproin as proposed by Lien and Nawar¹³⁹ is shown in Figure 11. Propane, propene, carbon monoxide, and carbon dioxide, which these workers previously identified in the heated triglyceride¹⁴¹ as well as in heated valine,¹¹⁸ were also present when both the amino acid and the triglyceride were heated together. Methane, ethane, ethene, butane, butene, pentane, pentene, hexanoic acid, and dicaproin, which were found to form in heated tricaproin, were also present in the heated mixture. Also, ammonia, acetone, isobutane, isobutylene, and isobutyraldehyde which were produced by

TABLE 16

**Decomposition Products Identified from Valine-tricaproin
Mixture Heated at 270°C for 1 hr**

Carbon monoxide	Isobutylene
Carbon dioxide	Pentane
Ammonia	Pentene
Methane	Acetone
Ethane	Isobutyraldehyde
Ethene	Hexanoic acid
Propane	Caproic nitrile
Propene	Caproic amide
Butane	<i>N</i> -Isobutylcaproic amide
Butene	Dicaproin
Isobutane	

From Lien, Y. C. and Nawar, W. W., *J. Food Sci.*, 39, 917, 1974. With permission.

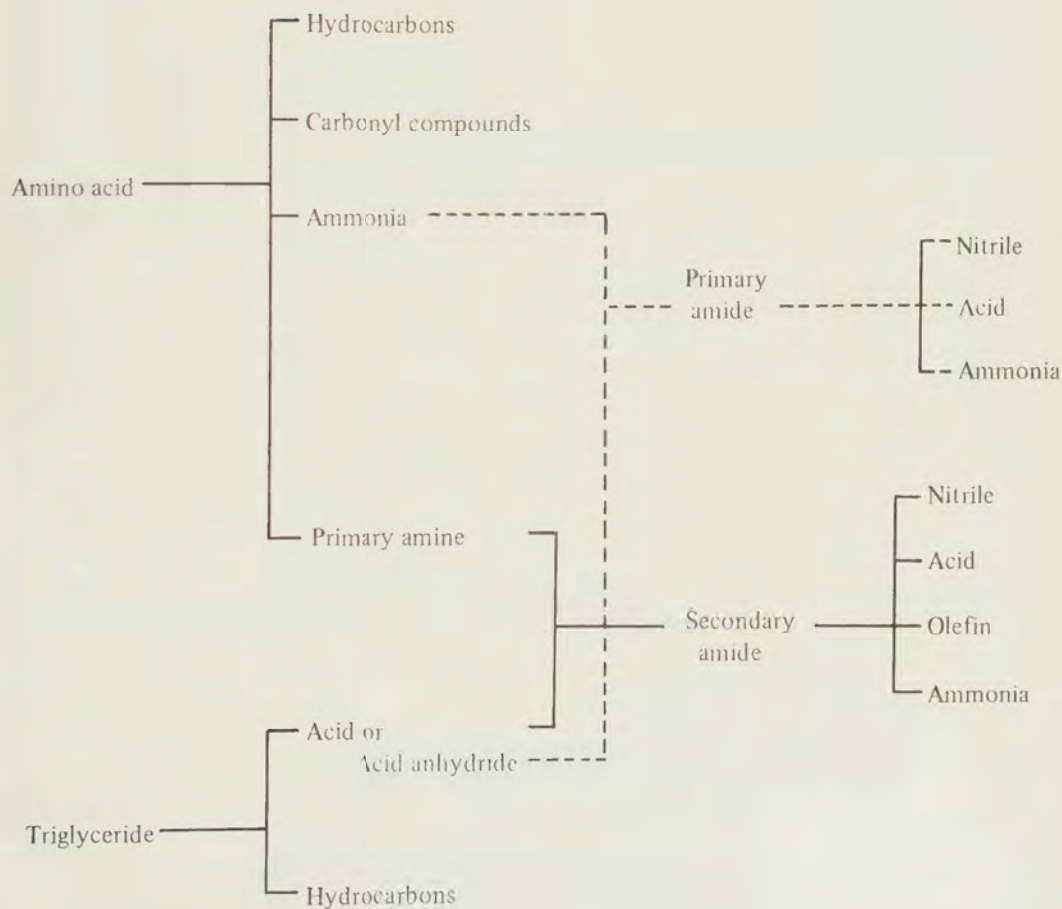


FIGURE 11. Scheme for interaction of amino acid and triglyceride upon heat treatment. (From Lien, Y. C. and Nawar, W. W., *J. Food Sci.*, 39, 917, 1974. With permission.)

heating the amino acid, valine, were individually present when both the amino acid and triglyceride were heated together. On the other hand, acrolein, methyl hexanoate, hexanal, 6-undecanone, 2-oxoheptyl hexanoate, propanediol dicaproate, propenediol dicaproate and oxopropanediol dicaproate (all observed in heated tricaproin), and isobutylamine, *N*-isobutylidene-isobutylamine, and diisobutylamine (all observed in heated valine), could not be detected when valine and tricaproin were heated together. In addition, some compounds not previously observed when the individual substrates were heated have been identified as interaction products formed during heating of the substrate mixture. These are caproic nitrile, caproic amide, and *N*-isobutylcaproic amide. It is evident from the work of Lien and Nawar^{118,119,139} that in a simple system containing only one amino acid and one triglyceride, substantial interaction does take place during heating. It is also quite evident that studies on the heat decomposition products of individual components using model systems can aid significantly in unraveling thermal interaction mechanisms in more complex mixtures.

A large number of thermal oxidation products of lipids are formed, in keeping with the size of the molecules, degree of unsaturation, and type of substitution. Thermal oxidation of lipids can occur at 60°C in the presence of a few free radicals, but most of the degradation occurs at 200 to 300°C. Lactones, alcohols, ketones, and lower fatty acids are associated with the thermal oxidation of lipids.

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FISH

Fish was preserved originally to insure adequate supply. Today the preservation of fish is geared to satisfy specific market demands. Various methods of fish preservation are employed: (1) pickling with salt crystals or salt solutions, (2) air or oven drying, (3) smoking, (4) marinating, and (5) sterilization (autoclaving) in tins using precooked fish packed in brine or olive oil.

The method of preservation influences the organoleptic characteristics of the finished product. Cod (Atlantic, *Gadus morhua* L.), salted and dried to about 40% moisture, is marketed under the name *bacalao*. Fish, such as haddock and cod, that is cured without salt by being split and hung in the air to dry is known commercially as stockfish. Herring, cod, salmon, sardine, and eel are usually smoked; eel and herring are also marinated. Boiled sardines, to which olive oil is usually added, are canned much like tuna of the species *Thunnus thynnus* (red) and *Thunnus alalunga* (white tuna). Salmon, mackerel, and other fish suitable for slicing are usually canned after preliminary boiling. The preparation of anchovy paste falls in the category of foodstuffs preserved by salting. Although flavor ingredients are seldom used as flavoring in the above preservation processes, a few spices, such as laurel, are used in marinating and sometimes in smoking fish.

VEGETABLES

*Pickled Vegetables**Salt-pickled Vegetables*

In general, preservation by covering the vegetable directly with salt (similar to the preservation of meat or fish) is not feasible. Salt-pickling is carried out by pickling the product in salted water. The percentage of salt in the pickling solution is expressed in degrees measured by a special hygrometer or salinometer, which is graduated to show the percentage of saturation of sodium chloride solution. The scale of the salinometer is divided into 100 degrees (1 to 100); one degree corresponds to 0.265% salt in the solution. The solution is saturated when it contains 26.5% pure sodium chloride.

Pickling is effected by using solutions with variable percentages of saturation depending on the final use. The preservative action of salt often is combined with the flavoring action of spices, aromatic herbs, and other flavor ingredients, especially if the pickled products are intended for direct consumption and not for use in subsequent food manufacturing operations. Specific methods of pickling may require the addition of specific flavor ingredients; for example, the addition of orange peels to the solution for home pickling of black olives is a fairly common practice.

An old recipe for the preparation of a flavored solution for generic home pickling of legumes follows:

*Pickling Solution for Legumes**Ingredients:*

Basil	100 g
Laurel leaves	10 g
Coriander	8 g
Mace	4 g
Juniper, minced	2 g
Salt	1 kg
Water	10 l

Preparation:

1. Mix all ingredients together and bring to a boil.
2. Allow the mixture to cool; strain.
3. Add legumes to pickling solution. The legumes will remain satisfactorily preserved for a few weeks.

Pickling is mainly used for preservation of vegetables in the fresh state. Therefore, unless resulting products with specific flavors are desired, pickled products in general are prepared to exhibit a flavor as close as possible to the original organoleptic characteristics.

Vinegar-pickled Vegetables

The preparation of vinegar-pickled vegetables is somewhat different and requires either the initial boiling of fresh vegetables or a preliminary salt pickling. If the vegetables have been initially salt-pickled, they should be washed thoroughly to remove the salt prior to vinegar pickling. Wine vinegar is highly desirable for pickling, although it may not always be available. If permitted by local regulations, vinegars from various sources and also acetic acid solutions are employed.

Flavoring of vinegar-pickled vegetables, although considered an indispensable operation in all cases, is extremely functional where vinegars other than wine vinegar are employed. A large number of

spices, herbs, seeds, fruits, roots, and legumes are used for flavoring vinegar-pickled vegetables. They are used individually or in blends. The flavor ingredients most commonly employed include white and black peppers, garlic, ginger, clove buds, cayenne pepper, coriander, nutmeg, chili, cinnamon, mustard seeds, horseradish, laurel, and paprika. Depending on the application, the whole spice, powder, oleoresins, or essential oils can be used. The flavor ingredients should be dispersed in the vinegar used for pickling.

Chutney

Chutney consists of a salad of comminuted vegetables and fruits in a rather sweet, thick sauce. The choice of vegetables and fruits, which is quite varied, includes onion, carrot, cauliflower, cucumber, beet, raisin, apricot, apple, etc. Chutney sauces contain a variety of products, although they primarily consist of sweet syrup, vinegar, soy, caramel, and several flavor ingredients, usually powdered seeds or fruits from ground spices. Powdered spice also contributes to the thickening of the sauce. Mace, pepper, paprika, and curry are frequently used together with wheat flour and natural gums. (For a further discussion of the flavor ingredients of chutneys, see p. 801.)

The vegetables and fruits are boiled for about 20 minutes together with water and sugar; then spices and salt are added. The mixture is again brought to boiling, then allowed to simmer for about 15 minutes. Vinegar and acetic acid are added, and the mixture is again brought to boiling; it is allowed to cool and then poured into glass containers together with a thick sauce prepared separately.

Oil-pickled Vegetables

Oil-pickled vegetables, as vinegar-pickled products, must undergo a preliminary cooking. Also vinegar-pickled products may be subsequently pickled in oil. The addition of flavor ingredients to oil-pickled vegetables is effected sometimes by two methods. In the first method flavor ingredients are added during the preliminary cooking of the vegetables; in the second, the flavors are added during preservation. In both cases the flavoring of oil-pickled vegetables is usually less intensive than vinegar-pickled products. Laurel is a flavor commonly employed during preservation; the flavoring consists of whole laurel leaves kept immersed in pickling oil. In addition to preserving vegetables, oil pickling is also frequently used to preserve fish.

Potatoes

Boiled and fried (chips) potatoes or dehydrated potato flakes exhibit different aromas because of the different degradation mechanisms involved as aroma precursors during cooking. Other ingredients, such as fats used for frying, obviously yield different flavor characteristics in the finished products.

Boiled Potatoes

The aroma of boiled potato is believed related to a Strecker's degradation and to the autoxidation of amino acids, sugars, and fats present in the tuber. Following are components reported present in boiled potato flavor:

Components of Boiled Potato Flavor

Alcohols		Sulfur-containing Compounds (continued)
Ethyl alcohol		Diethyl sulfide
Methyl alcohol		Hydrogen sulfide
		Methyl ethyl disulfide
		Methyl ethyl sulfide
		Methyl isopropyl disulfide
		Methyl <i>n</i> -propyl sulfide
		2-Propyl mercaptan
		<i>n</i> -Propyl mercaptan
Carbonyl Compounds		Ketones
Acetaldehyde		Acetone
Acetone		2-Butanone
2,4-Decadien-1-al (50%)		2-Heptanone
2-Decen-1-al		2-Hexanone
2-Methylbutanal		2-Nonanone
3-Methylbutanal		2-Octanone
2-Methylpropanal		2-Pentanone
Nonanal		
2-Nonen-1-al		
Propanal		
Propenal		
2-Undecen-1-al		
Sulfur-containing Compounds		Pyrazine Derivatives
Dimethylsulfide	} 90%	Decomposition products of methionine
Methyl mercaptan		2,5-Dimethylpiperazine
Dimethylsulfide	} 70%	
Ethyl mercaptan		

Fried Potatoes

The flavor of fried potatoes is derived mainly from the autoxidation of the fats and oils used for cooking rather than the action of heat on the potato. Potato aroma is rather unstable and reverts readily from pleasant to unpleasant on aging. Aging causes a decrease of the dienal content with a corresponding increase of carbonyl compounds (following the darkening reaction) together with a characteristic stale odor. The aroma of freshly fried potatoes has been reported caused by the following components:

Aldehydes in Fried Potato
Aroma

Heptanal	Octanal
2-Hepten-1-al	2-Octen-1-al
Hexanal	Pentanal
2-Hexen-1-al	

Dehydrated Potato Flakes

The aroma of this product is characterized by the presence of carbonyl compounds derived from the same precursors. The carbonyl compounds are formed as degradation products of saturated and unsaturated fatty acids. These are formed by mechanisms similar to those indicated for boiled potatoes with the added effects of a heat history from the dehydration process. The off-flavors developed during flake manufacture can be controlled in part by using antioxidants.

MISCELLANEOUS PRODUCTS

Tobaccos

The various species of *Nicotiana* (*N. tabacum*, *N. rustica*, *N. petunioides*, *N. glauca*) belong to the family *Solanaceae*. Among these the countless varieties of *N. tabacum* and *N. rustica* represent the most important source for both smoking and sniffing tobaccos. *Nicotiana* plants are native to the Americas; the first seeds were carried to Europe by Cortez in 1519. Because of the widespread *Nicotiana* cultivations throughout the five continents, tobacco varieties are classified according to the geographical name of the growing site—America, Asia, Australia, and Europe. The most important varieties are listed below:

1. American tobaccos: Kentucky (light and heavy)
Virginia (yellow, sweet, and dark, heavy)
Maryland (yellow, sweet, aromatic)
Seed leaf (with silky leaves, sweet)
Havana (highly aromatic and sweet)
2. Asian tobaccos: Middle East or Turkey
3. Izmirn tobaccos: Sansum and Bafia
4. Australian tobaccos: Sumatra and the Philippines
5. European tobaccos: Produced in the Netherlands, Belgium, Germany, Russia, Hungary, and Italy. They are used in cigarette, cigar, pipe, sniffing, or chewing tobaccos; the latter is steadily declining in use.

Tobacco leaves are treated for their specific end-use. Sniffing tobaccos are made from the most aromatic leaves, which are comminuted, fermented, and flavored to enhance the aroma. Chewing tobaccos, consisting of pressed plaits made from tobacco leaves, are treated with sugar and only slightly flavored. Smoking tobaccos are manufactured using selected qualities most suitable for the specific end-use (e.g., cigarette, pipe, cigar). The dried leaves, free of the woody portion, are used as the outer wrapping for cigars or are finely comminuted for use in pipe and cigarette tobacco blends.

The characteristic aroma of the properly selected tobacco varieties, enhanced and modified by the specific manufacturing process, is considered the most important factor in the finished product because it directs the specific preference of consumers. Natural tobacco flavor may not be extremely palatable because of its constituents and may require additional flavoring. However, the flavoring of tobacco is often misconstrued as a means to mask the natural aroma of less valuable tobacco qualities. Therefore, cigarette manufacturers tend to shy away from claiming additional flavoring in product brands.

Tobacco flavoring may have a twofold function: (1) flavor enhancement and/or improvement, and (2) flavor modification. Flavor enhancement is obtained by the addition of natural tobacco extracts, whereas flavor modification is obtained by the use of "blenders." By extracting the tougher, woody portion of leaves and stems of the best tobacco qualities with water-alcohol or water-ether mixtures, natural tobacco extracts are prepared; these are used to improve the aroma of less valuable tobaccos. Havana tobacco extracts, for instance, are useful in the processing of Puerto Rican tobaccos. Blenders, or finishing flavors, are used to enhance those pleasant flavor notes and to soften the harsh notes. These flavor ingredients are products from various plant sources and make up a substantial portion (up to one-fifth by weight) of the finished product.

During softening of tobacco leaves by conditioning under controlled humidity conditions, the humidified flexible leaves are best suited for the addition of flavors. Specific flavor ingredients, dispersed in liquid aqueous sweetener or solutions (honey, molasses, apple and plum juices, fig tincture, sugar, cocoa, licorice, etc.), are brought to boiling, and the tobacco leaves (previously conditioned with steam) are dipped in the solution. The dip is usually quick and is followed by draining, stretching, drying, and rehumidifying of the leaves.

Each tobacco quality requires specific complementary flavoring and a more or less pronounced sweetening. This at times is carried out by using wine, rum, propylene glycol, alcohol, and glycerol as the dispersing phase for resinoids, essential oils, fluid extracts, and isolates. The most frequently

encountered flavor ingredients include storax and opopanax resinoid, Tolu and Peru balsams, absolutes or extracts (melilotus, liatris, tonka bean, vanilla), valerian tincture, orris tincture, and the essential oils of geranium, rose, cinnamon, clove, etc. The essential oils must be terpeneless. Isolates and synthetic ingredients are used at minimum levels and only for marginal flavoring. For example, a few aldehydes enhance the floral headnote. Government regulations (which vary from country to country) provide excellent guidelines as to which products may be used in the formulation of tobacco flavors.

Sniffing tobacco draws its aroma from fermentation of the leaves. Tobacco for cigars is only slightly flavored, cigarette and pipe tobaccos draw the characteristic aroma from selected tobacco qualities (Virginia, Carolina, Burley, Lalaika, and Turkish tobaccos) and from added flavor ingredients and sweeteners.

The formulation of tobacco flavors follows specific criteria entirely different from the flavoring criteria used in food and perfumery. The aroma of the flavor ingredients develops in two stages, *i.e.*, before and after combustion. It is impossible to give fixed guidelines with very few exceptions. Sometimes fruit flavor imitations, such as pear, apple, and peach, may be required to strengthen certain flavor notes. More frequently honey flavor imitations are used. Rum flavor, widely employed in the manufacture of cigars, is usually an imitation prepared by guidelines similar to those used for the preparation of rums for liqueur formulations; these usually contain rum ether.

Chewing Gums

The advent of the chewing gum industry is fairly recent—about one hundred years old. The basic raw material consists of chicle gum (the latex of naseberry or sapodilla) native to Yucatan and Central America. Two basic groups of gum products are manufactured: (1) the so-called chewing gums (in sticks or bonbons) and (2) bubble gums. For the individual products the gum is prepared in standard bases by specialized manufacturers who supply, together with the gum base, recommendations for its use. The gum is employed in amounts varying between 17–30%, depending on the product type and the blend of gums used. Other ingredients present in varying amounts include sucrose (46–59%), glucose (18–23%), water, glycerol, flavor ingredients, and colorants.

Manufacture consists of mixing the ingredients in a special mixer equipped for heating the blend to 60°C; then the mixture is kneaded by passing the plastic dough through cylinders, and finally the mixture is cut into sticks, balls, etc. The gum is heated in the mixer and softened without melting; then glucose, sucrose, water, glycerol, flavor ingredients, and colorants (optional) are added in this order. The homogenous dough is heated to 45°C, and the mass is poured and cut into loaves that are allowed to cool. The loaves are then rolled or stamped, and the manufacturing cycle is terminated by additional operations suitable to prepare the desired end product. The various operations, briefly described above, require special equipment and skill; the discussion of the latter, however, goes beyond the scope of the book.

A number of precautions are required that, in ultimate analysis, characterize the chewing gum production as to smoothness, hardness, plasticity, cohesiveness, lack of tackiness, elasticity, and flavor. The original chewing-gum product was almost flavorless; then it was made sweet and finally aromatic. Today effective flavoring is at the base of most chewing- and bubble-gum production. Therefore, suitable flavor ingredients with specific requirements must be employed. Chewing gum flavor ingredients must be insoluble in water but readily soluble in the gum. This minimizes flavor loss by solubility in saliva and insures the gum acting as a long-lasting flavor carrier. The flavors must also act as plasticizers in the gum and exhibit heat stability throughout the manufacturing cycle. Raw or rectified essential oils are well suited for this application. The most popular chewing-gum flavors include spearmint, peppermint, anise, wintergreen, cinnamon, tutti-frutti, cherry, and banana.

Drug Flavors

The original production of drugs was based on the extraction of herbs. In addition to the "active curative principles," the botanical source contained flavor ingredients that often imparted palatability to the drug extract. By blending various herb extracts, it was often not too difficult to formulate products having sufficiently agreeable odor and flavor for consumption.

With the increasing use of synthetic drugs and drug principles of animal origin, the number of drugs with extremely disagreeable odor and flavor has increased. This, in turn, has greatly increased the need for flavors capable of masking drugs, especially those presented in liquid form for oral consumption. Although the production of drugs today is increasingly oriented toward the manufacture of coated tablets, capsules, injectable specialty products, and suppositories, the formulation of liquid products (syrups and drops) for direct oral consumption together with pills and granulated products still represents a large percentage of drugs requiring suitable flavoring. "Medicines" are often associated with products having exclusively unpleasant odor and flavor. For many years no effort had been made to render more acceptable or palatable the odor or flavor of drugs. Today the trend has reversed; even products for external use, such as pastes, pomades, and unguents, are suitably formulated with agreeable fragrances. Although not all drug principles have disagreeable odor and flavor, some exhibit nauseating aroma characteristics.

The flavoring of solid drugs (tablets, etc.) may be readily attained by hard coating with sugar-based flavors, while the flavoring of liquid products is much more complicated. The simple addition of sugar or other sweeteners to liquids may not only be insufficient but may also produce detrimental effects. The criteria of sweetening the flavor of drugs because of their bitter or salty flavor is absolutely incorrect. The formulation of "medicinal" syrups is perfectly valid as long as certain basic criteria are followed, since the addition of sweeteners to certain products may yield totally unpalatable flavors. On the other hand, flavor modifications may yield unexpected and extremely interesting results. A product with a very bitter taste is a case in point. Theoretically the dispersion of the bitter active principle in the syrup should be sufficient to overcome the bitter flavor; in practice the addition of a few bitter flavor ingredients yields a new bitter note that yields a very palatable flavor sensation with only a minimum addition of sweetener. The addition of fruit flavors and sugar to a drug is not adequate to mask the original unpleasant flavor. For example, sweetening a bitter note and then adding a strawberry, currant, or raspberry flavor may yield totally unacceptable flavor results, whereas shifting the original bitter note to a bitter vermouthe note together with the addition of a slight citrus note may yield an excellent solution to the problem. A few exceptions should be mentioned, however, such as drug products for children, who show a marked preference for fruit flavors.

A preliminary, thorough study of the original drug odor and flavor to identify the most suitable modifications for the creation of a new flavor without attempting to transform it into a fruit-flavored product is highly recommended. Following this preliminary investigation and taking into account the expected use levels of the product, the chemical constitution, and the carrier, the task of compounding can be undertaken by selecting the flavor ingredients from broad categories, such as fluid extracts, soluble essential oils, isolates, synthetic products, and concentrated and lyophilized fruit juices. Compounded flavors used for syrups, liqueurs, or pastry should be considered first.

In the choice of flavor ingredients, the chemical composition of the drug must be taken into consideration, since any potential reaction between flavor ingredients and the active drug principles must be avoided. Once any potential reaction between the various ingredients is avoided and the desired flavor results are attained, it is still necessary to control the activity of the drug by suitable analytical means over an extended period of time; certain reactions can develop at an extremely slow rate, and the resulting effects are often unpredictable. As a result of several investigations conducted in France at the Department of Pharmacy of the University of Grenoble, analytical techniques have been developed to monitor both flavor retention and residual pharmacological activity over a period of time in drug preparations. A few flavor houses have developed a series of flavor ingredients specifically suitable for effective drug flavoring, basing their developments on previous experience with typical drug formulations. These flavor ingredients are highly stable and nonreactive toward known drug active principles.

Aside from the preference for fruit flavors by children in general, drug products are commonly flavored with the following notes:

1. Fruit notes: raspberry, strawberry, currant, cherry, peach, banana, plum, orange, lemon, citron, and mandarin
2. Variations of bitter notes: cinchona, rhubarb, gentian, vermouthe, and bitter orange
3. Masking flavors with high yields: anise, peppermint, licorice, vanilla, cocoa, and caramel.

Finally, it should be emphasized that in many countries the approval of flavor ingredient for use in food does not include its use for flavoring drug products. For example, current U.S. regulations require that the complete drug formulation, including active principle(s), inert fillers, colorants, and flavor ingredients, be thoroughly tested for pharmacological activity and toxicological safety. The objective is to prevent potential toxicological hazards and/or losses of pharmacological activity resulting from unexpected chemical and physicochemical interactions between the drug principle and other components in the formulation. Similar regulations are followed in West Germany and in Italy, but many other European countries do not regulate the specific use of approved flavor ingredients in drug products.

Animal Feeds

It is impossible to determine exactly whether animals in general or individual animal species in particular experience similar flavor and olfactory sensations to humans. It has been proven, however, that the olfactory sensation is in general much more developed in animals than in humans. A few animal species prefer certain specifically flavored foods, and they clearly show a repulsion for others. Agreeable aromas stimulate similar physiological reactions in animals and humans. It is apparent that flavor ingredients can have a functional use in solving certain animal nutrition problems, for it is well known that palatable food is digested much easier than less appetizing food. This in turn causes feed to be assimilated by animals at a much faster rate than less aromatic feeds. Thus flavors add extra nutritional value to feeds. The efficient consumption of feed has a great economic importance in large-scale farming of domestic animals. The larger the amount of feed ingested and digested by the animal, the higher the yields of meat from poultry and cattle, eggs from poultry, and milk from cows.

The creation of animal feed flavors was developed and conditioned by observations of the alimentary habits of various animals based on species and age. Hogs are fed a sugar-flavored feed, whereas mild flavors are added to the feed of suckling pigs. Cattle generally show a preference for anise- and fennel-flavored feed together with sweet, sugar-like flavors. Calves' feed is flavored with milk flavors, whereas the molasses aroma is particularly preferred by more adult animals. Cattle feed often contains preservatives and antioxidants that contribute to the preservation of carotene and vitamin E content in the feed. However, a few preservatives tend to destroy rather than preserve the carotene content; for example, a decrease in carotene was observed in animal feed treated with zinc-bacitracin and sodium metabisulfite.

From experiments conducted on dogs, it was observed that the addition of beef-bone flavors favored ingestion of several different foods. Similarly, food flavored with products somewhat undefinable by human taste standards favored the ingestion of feed by chickens, pheasants, etc. Although young animals in general (swine and cattle) show preference for those flavors imitating the flavor of the mother's milk, the type of feed selected and therefore its intrinsic flavor characteristics are also important in the formulation of complementary flavors.

Although it is possible to attain quite satisfactory flavor results using milk flavors, optimum flavoring is achieved with milk flavor formulations specifically tailored for swine or cattle. These flavors must be compounded independently of their specific organoleptic characteristics for optimum masking and blending effects and in combination with the odor and flavor of the ingredients selected for the feed. Flavoring difficulties are greatly increased in special feeds containing anti-stress ingredients, vitamins, and medicines. These require the inclusion of active flavor principles with characteristic notes capable of masking. Pelleted feed requires the use of heat-resistant flavor ingredients for manufacturing purposes. It is important to have available both powdered and liquid flavor ingredients with equivalent aroma yields for both effective surface and mass flavoring of the feed.

The above information may be summarized as follows:

1. Molasses notes are particularly suitable for flavoring cattle feed.
2. Fruit notes in general are preferred by horses.
3. Milk-flavored feed, completed by cocoa notes and similar variations, are particularly attractive to swine.
4. Cocoa flavors, complemented by milk and wintergreen notes, are particularly suitable for flavoring cattle feed.

5. The flavoring of poultry feed is more complex, as experiments conducted in several parts of the world have yielded conflicting results. Together with the mentioned undefinable "human" note, the smoky note appears to have more positive effects.

6. Anise notes are also of interest and can be suitably modified for flavoring rabbit feed.

Feed flavors are being marketed with increasing frequency in powder form (plated over carriers or spray-dried). Such powders very often contain sweeteners in addition to the principle flavor ingredients. The addition of flavors to bulk feed is readily attained by using suitable dispersing agents (surfactants) or by purely mechanical means.

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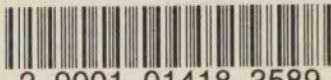












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