Meat Flavor: Lamb

IRWIN HORNSTEIN and PATRICK F. CROWE

Eastern Utilization Research and Development Division, U. S. Department of Agriculture, Beltsville, Md.

The characteristic aroma of heated lamb is obtained from the fat. A major portion of the odor, obtained upon heating, is contributed by carbonyl compounds. These flavor compounds, or their precursors, are apparently present in only trace amounts in the fat. Heating the bulk of the triglycerides, after removal of these trace components, does not generate the characteristic lamb aroma. The lean meat portions contribute a basic meaty flavor similar to that obtained from lean beef and lean pork. Volatile compounds isolated from the lean lamb as well as the physical data obtained for compounds not completely characterized are similar to those obtained from lean beef and pork. The flavor precursors in the lean lamb are low molecular weight, water-soluble compounds that produce characteristic meaty aromas on heating.

THE PRESENT INVESTIGATION was undertaken as part of a continuing program on studies of flavor in meat. Results obtained in the study of lamb flavor are presented and compared with results previously obtained in flavor studies on beef and pork (*6*, *7*). We have, as in previous work, equated aroma with flavor on the assumption that aroma is the major contributor to flavor. Techniques previously developed for the study of flavor in pork and beef have constituted a substantial portion of the methodology used in the study of lamb flavor. Particular emphasis has been placed on the contribution of lamb fat to aroma. A major purpose of this study was to learn if the generalizations made concerning beef and pork flavor, i.e., that the lean portions of the meat contribute a similar meaty flavor while the fat portions contribute the flavor differences in these meats, could be extended to lamb.

Experimental

Lean lamb was obtained from four carcasses purchased on the open market. Lamb fat was a 50-pound composite sample of kidney fat obtained from a number of animals. Lean and fat were stored separately at 0° F. until needed. The lean was not studied muscle by muscle, but rather as a composite, since the authors' previous work on beef and pork had shown that the flavor and contributions from different muscles were identical. Lamb kidney fat was used for all fat studies.

Lean Lamb. A freeze-dried lamb powder extract containing the flavor precursors was prepared as follows: Lean meat was thawed, fat and connective tissue were removed, and the trimmed meat was ground in an electric grinder maintained at 32° F. The ground lean was blended with ice-cold, distilled water. The slurry was centrifuged at 32° F. and the extract filtered under suction through a thin layer of Filter-aid to remove solidified fat particles. Lyophilization of this extract yielded a powder approximately 2.0% by weight of trimmed meat. This lamb powder was heated at 100° C. under vacuum in a specially designed, all-glass apparatus (*7*), and the total volatiles were condensed at liquid nitrogen temperature. Pumping was then stopped, but the vacuum was maintained by the closing of properly placed stopcocks. The trap containing the total condensate was allowed to come to room temperature, and spontaneous distillation took place. The most volatile fraction was collected at liquid nitrogen temperature; a second fraction, mainly water, was collected at dry ice–isopropanol temperature; and the highest boiling fraction remained in the original trap. The most volatile fraction (I) and the least volatile fraction (II) were analyzed. The contents of the trap containing water were not studied. Methods and apparatus are described in previous papers (*6*, *7*).

The more volatile fraction (I) was analyzed for acidic, basic, and carbonyl compounds. Carbonyl compounds were converted to their 2,4-dinitrophenylhydrazones by the addition to the cold trap containing fraction I of a 2*N* hydrochloric acid solution saturated with DNPH. In a separate experiment, the acidic compounds in I were converted to their ammonium salts by the addition to the cold trap, in which I was collected, of 5 ml. of 5% ammonium hydroxide. In similar fashion, basic compounds were converted to their hydrochlorides in a separate experiment by the addition of 5 ml. of 2*N* hydrochloric acid (*7*). The basic compounds in the 2*N* hydrochloric acid solution were recovered as their hydrochlorides by lyophilization of the solution. Addition of one drop of sodium hydroxide solution to a crystal produced the odor of ammonia. Paper chromatography, with methanol as the solvent, yielded no other aliphatic primary amines (*7*). Acidic compounds trapped in ammonium hydroxide were evolved by the addition of 2*N* hydrochloric acid; the major constituent was carbon dioxide. The fraction was analyzed for free sulfide ion by a modified procedure (*7*) of Marbach and Doty (*9*). Carbonyl compounds were analyzed as their DNPH's, using paper chromatographic procedures of Gaddis and Ellis (*2–4*).

The less volatile of the two fractions was a viscous, water-soluble material. Approximately 100 mg. were obtained from 30 grams of dried powder. The pH of a 1% solution in water was approximately 3.8. The infrared spectrum of a film of this material gave major peaks at 3.15, 5.81, 6.32, 7.12, 8.9, 9.6, and 11.7 microns. The ultraviolet spectrum of 2 mg. per ml. of water had a maximum at 290 to 295 mμ. This fraction was separated on paper by butanol saturated with water to give three bands at R_F values corresponding to those previously obtained from beef and pork (*6*). Lactic acid was determined by the method of Hullin and Noble (*8*).

Precursors of lean lamb flavor were isolated from the lyophilized powder by gel filtration, on Sephadex G-25, of a 25% solution of the powder in water (*10*); the eluant was 0.005*M* NaCl. The high molecular weight fraction, which was eluted first, was lyophilized to yield a brown powder similar in appearance to the unfractionated powder. The low molecular weight fraction was also lyophilized to yield a white fluffy powder. The ratio of high

to low molecular weight fractions was about 2:1.

Lamb fat. Lamb fat rendered under nitrogen was heated for 4 hours at 110° to 115° C. in an all-glass apparatus. The heating chamber was 5.5 cm. in diameter and 14.5 cm. deep, equipped with 55/50 ⅋ joint and appropriate connection for gas inflow and exhaust. To remove any possible carbonyl or acid contamination from the sweep gases, the air or nitrogen was purified by bubbling through a saturated solution of DNPH in 2*N* phosphoric acid to remove carbonyl compounds, then through a tower containing sodium carbonate and silica gel for the removal of acidic components, and finally through a silica gel drying tower. The purified gas at a flow rate of 3 to 5 liters per hour was passed across the surface of the heated fat and into a trap cooled in dry ice-isopropanol or in liquid nitrogen. Both volatiles and residual fat were analyzed for carbonyl compounds and for free fatty acids.

Carbonyl compounds in the lamb fat volatiles were converted to their DNPH's by adding a saturated solution of DNPH in 2*N* hydrochloric acid to the contents of the cold trap and allowing the mixture to stand overnight. Total volatile carbonyls were estimated by measuring the ultraviolet absorption of the DNPH's at 349 to 352 mμ, the region of maximum absorption for this mixture; and monocarbonyls were determined by the paper chromatographic procedure of Gaddis and Ellis (*3*). Carbonyls present in the residual fat were converted to DNPH's by the method of Schwartz *et al.* (*11*, *12*). The mixture was separated and monocarbonyls identified by a combination of these methods. Briefly, a 5 to 10% solution of lamb fat in carbonyl-free hexane was slowly percolated through a 10-gram celite column impregnated with DNPH, water, and phosphoric acid (*12*). The DNPH's formed were eluted along with the hexane and fat; elution was completed by washing with additional hexane. Highly polar carbonyls remained on this column. The hexane eluate was then chromatographed on a 1:1 w./w. mixture of Seasorb (MgO) and Celite 545 (*11*). DNPH's and unreacted DNPH were held to the column, and the hexane eluate contained essentially all of the fat. Removal of the hexane under vacuum from the carbonyl-free eluate left more than 97% of the original sample. The monocarbonyls and most of the dicarbonyls were eluted from the column with a mixture of 25% nitromethane: 75% $CHCl_3$ v./v. (*11*). The solvent was removed from the eluate and the DNPH's separated into groups by chromatography on partially hydrated alumina. Carbonyls of increasing polarity were successively eluted with a 1:1 mixture of hexane-benzene, benzene, and ethanol. The aliphatic aldehyde fraction eluted by hexane-benzene was further separated by paper chromatography (*2–4*). Ultraviolet absorption at 352 mμ was taken as a measure of the carbonyl content of each fraction (*3*).

Free fatty acids were determined by first separating them from the fat on a basic ion exchange resin, methylating these acids, and finally separating and determining the esters by gas chromatography (*5*).

Results and Discussion

As in beef and pork, the precursors in the lean lamb were extractable with cold water. Heated lamb patties, prepared from water-extracted, ground, lean lamb, were tasteless, while patties prepared from extracted meat, reconstituted with the extract, were characterized as meaty but with no area of agreement as to the kind of meat utilized. Two volatile fractions were obtained by the vacuum pyrolysis and subsequent fractionation of the lyophilized water extract of the lean. The less volatile of the two fractions had a pleasant, fruit-like aroma that, on standing, assumed a desirable meaty aroma. The aroma characteristics were identical to those from the equivalent fractions previously obtained from beef and pork. The infrared and ultraviolet spectrophotometric data, as well as the chromatographic separations for this fraction, were also identical to those reported for the equivalent beef and pork fractions (*2*). Lactic acid again constituted about 90% of this fraction.

The more volatile fraction was characterized by a strong ammoniacal and a weak sulfide odor. The latter was in contrast to the strong sulfide odor present in the equivalent pork and beef fractions. Thus, lamb evolved 2.5 mg. of ammonia per gram of powder and only trace amounts of hydrogen sulfide, while beef and pork yielded about 1.7 mg. per gram of ammonia and 0.1 mg. per gram of hydrogen sulfide. The pH of a 10% solution of lamb powder in water prior to heating was 6.0 compared to values of approximately 5.2 for similar solutions of beef and pork powder. The differences observed in the yields of ammonia and hydrogen sulfide may well be related to this pH difference. To be certain that the pH was indeed 6.0, and that the ammonia evolved was as high as shown, and that hydrogen sulfide was present only in trace amounts, these experiments were repeated on a carcass purchased several weeks later and on the carcass of another animal whose entire history prior to, and after, slaughter was known. In both instances, the pH of the lamb powder was 6.0 ± 0.1, the ammonia was evolved in the order of 2.5 mg. per gram of powder, and the amount of hydrogen sulfide obtained was just detectable. The major carbonyl compound isolated was acetaldehyde; smaller amounts of formaldehyde and acetone were also obtained for beef and pork (*6*).

The high and low molecular weight fractions obtained from the lean lamb water extracts after gel filtration were carried through the vacuum pyrolysis procedure and the volatiles trapped and fractionated. No appreciable aromas or volatiles were obtained from the protein fraction, but the low molecular weight fraction yielded the aromas and compounds associated with the unfractionated powder. This too was similar to results obtained from beef and pork where the separation into high and low molecular weight fractions was obtained by dialysis rather than gel filtration (*6*), and is in agreement with the observation of Batzer *et al.* regarding beef precursors (*1*). Lamb fat, heated in an air stream or under nitrogen, developed an aroma usually associated with mutton. The odor was strongest in samples in which small amounts of water were added. Preliminary examination of the volatiles for carbonyl compounds and free fatty acids was negative. To check this observation, lamb fat plus water was heated in air under reflux for 4 hours, transferred to the high vacuum apparatus, heated to 100° C., and the total volatiles condensed at liquid nitrogen temperature. A very strong, mutton aroma was obtained. Water was added to the condensate and the slightly opalescent solution divided into three 3-ml. aliquots. To the first tube, 2 ml. of distilled water were added; to the second, 2 ml. of a 2*N* solution of phosphoric acid; and to the third, 2 ml. of 2*N* phosphoric acid saturated with DNPH. The glass-stoppered tubes were shaken and at intervals the odors noted. The mutton odor remained in the tubes containing water and phosphoric acid, but there was a steady decline in odor intensity in the DNPH solution. However, even after 24 hours, an aroma reminiscent of mutton, but far less intense and of a different character, was still present. Spectrophotometric measurements indicated the presence of only trace amounts of carbonyl compounds; and analyses for monocarbonyl alkanals, 2-enals, and 2,4-dienals were negative. Since the analytical method used can detect less than a microgram of carbonyl, then the portion of lamb flavor attributable to carbonyl compounds is apparently present only in fractional p.p.m.

To learn if carbonyl compounds initially present in the fat were responsible for the aroma, rendered lamb fat was dissolved in hexane, and carbonyl compounds were removed as their DNPH's on a magnesium oxide column. Heating this carbonyl-free

Table I. Monocarbonyl Compounds Isolated from Lamb Fat

(Figures are in per cent of total monocarbonyls found)

Carbonyl	Unheated Fat	Heated in Nitrogen	Heated in Air
Saturated C_6	10	10	...
Saturated C_9	...	...	trace
Saturated C_{11}	...	...	trace
Saturated C_{16}	40	35	40
Saturated C_{18}	50	55	55
2-Enals			5[a]
Total μmoles of carbonyl in 10 grams of fat	0.94	2.23	1.08

[a] Equal amounts of C_9, C_{10}, and C_{11}.

Table II. Free Fatty Acids in Unheated and Heated Lamb Fat

Acid	Mg. per Gram of Free Fatty Acid per Gram of Fat		Per Cent of Total Free Fatty Acids	
	Before heating	After heating	Before heating	After heating
Linoleic	0.017	0.02	0.51	0.56
Oleic	1.49	1.91	48.26	48.05
Stearic	0.92	1.14	29.73	29.01
Palmitoleic	0.01	0.009	0.22	0.21
Palmitic	0.65	0.87	20.95	21.70
Tetradecadienoic	0.001	0.001	0.03	0.03
Tetradecenoic	0.001	0.001	0.03	0.03
Myristic	0.01	0.012	0.23	0.30
Lauric	0.0004	0.003	0.01	0.06
Decadienoic	0.0007	0.001	0.02	0.03
Decaenoic	0.0003	0.0008	0.01	0.02
Total mg. per gram	3.10	3.97		

fat in air did not produce the characteristic mutton-like volatiles, and the odor intensity was low. This would indicate that at least a high proportion of mutton aroma is carbonyl in nature or that the mutton aroma constituents or precursors are highly polar and are removed from the fat by the isolation techniques used. In any event, the odor compounds or their precursors are present in only small amounts in the fat, and the aroma is not continuously generated by heating the bulk of the triglycerides present in the fat.

Since carbonyls were apparently important contributors to lamb flavor but were not present in high enough concentration to be determined in the volatiles, the fat itself was analyzed for carbonyl compounds by the Schwartz method which frees bound carbonyls (*11*). Table I lists the monocarbonyls. The major carbonyls found, C_{14}, C_{16}, and C_{18}, are not ordinarily volatile and do not contribute to aroma; neither hexanal nor the small amounts of 2-enals obtained have a mutton aroma. Total carbonyl compounds as measured by ultraviolet absorption were considerably greater than those isolated from the monocarbonyl fraction. For example, prior to heating in air, approximately 14 μmoles of carbonyl calculated as saturated aldehyde per 10 grams of lamb fat were present. After heating in air or in nitrogen, this increased to approximately 25 μmoles of carbonyl per 10 grams of fat. The aliphatic aldehyde content prior to and after heating in air was about 1 μmole per 10 grams of fat and about 2 μmole after heating in nitrogen, and only this fraction has been accurately analyzed. These carbonyls are assumed to be responsible to a great extent for the mutton aroma. This paucity of volatile saturated and unsaturated carbonyl compounds isolated from lamb fat is in sharp contrast to the volatile monocarbonyls isolated from beef and pork fat where alkanals, 2-enals, and 2,4-dienals were isolated (*6*).

No free fatty acids were found in the volatiles from lamb fat. The free fatty acids found in the fat before and after heating are given in Table II. The results again are quite different from those obtained from pork and beef fat. The absolute amounts are lower. Thus, after heating, the free fatty acids in lamb fat totalled only 0.40%, this in contrast to values of 3.7% and 5.5% in beef and pork, respectively (*6*). In addition, the three major acids in lamb fat (oleic, stearic, and palmitic) constitute 99% of the free fatty acids. In beef and pork fat, these were also the three major free fatty acids, but they accounted for 80 to 85% of the free acids. These results may explain the low concentration of 2-enals and the absence of 2,4-dienals in heated lamb fat, since unsaturated free fatty acids may be oxidized more readily than the same acids present in the triglyceride.

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