

Lipid Degradation of Beef Stew with and without Vegetables

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Abstract

Stews were prepared by 2 processes and 4 treatments, and stored for 3 different storage periods. The two processes were beef cooked in a stew and stored in a polyethylene container at 5 °C (P1) and in a barrier bag at 0 °C (P2). The four treatments were beef cooked alone (T1), with onions (T2), with carrots (T3) and with onions and carrots (T4). Stews in P1 were stored for 0, 2 and 4 days and stews in P2 were stored for 0, 2 and 4 weeks. Cooking decreased the cephalin content by 39%, the lecithin content by 21% and most of the phospholipid fatty acid concentrations as well as the fatty aldehyde levels in the phospholipids of beef from stew. Process or storage did not significantly affect the level of either phospholipids, however cooking beef with carrots seemed to exhibit some protection against hydrolysis of cephalin. P1 stews had a higher TBA-value ($p < 0.05$) than P2 stews, and the TBA-value of P1 stews increased linearly during 4 days storage. The TBA-value was not affected ($p < 0.05$) by treatment for any of the stews and did not change significantly during 4 weeks storage in P2 stews.

Key words: beef stew, phospholipid, TBA value

Introduction

Today, use of foods cooked at a central location and put into barrier type bags with subsequent distribution to cafeterias, hospitals and fast food outlets is increasing. The use of barrier bags increases shelf life and retards flavor deterioration because it reduces lipid oxidation in products containing meat. Beef stews containing vegetables, some of which have been reported to inhibit lipid oxidation, can be cooked and then stored in barrier type bags. No studies were found, however, that investigated beef lipid degradation during processing and storage of beef stew in barrier bags or even in the conventional manner. Information concerning degradation of lipid during processing and storage in such products as beef stew is needed for a better understanding of the flavor deterioration. The major flavor deterioration in products containing meat is oxidative rancidity often called warmed over flavor (WOF). WOF develops within a few hours in refrigerated cooked meats and its primary cause is lipid oxidation⁽¹⁾. The major lipid components of the cell membranes of the meat are phospholipids, but they are only small part of the total meat lipid content.

Wilson *et al.*⁽²⁾ showed that 3.56% of total lipid in beef muscle containing 14.79% of lipid was phospholipids. Although phospholipids in beef muscle are present in such small amounts, they are the major lipids involved in the development of WOF⁽²⁻⁴⁾. Gokalp *et al.*⁽⁵⁾ showed that the neutral lipid fraction in beef had higher ($p < 0.01$) total percentage of saturated fatty acids and 27.6% more monounsaturated fatty acids than the phospholipid fraction in beef whereas the phospholipid fraction had significantly higher contents of dienoic, trienoic and tetraenoic acids. Each additional double bond in a fatty acid increases the rate of oxidation by factor of two⁽²⁾. The thiobarbituric acid (TBA) test is one of the commonly used chemical method for determining oxidative rancidity in cooked meat⁽⁶⁾. The TBA test measures the contents of secondary lipid oxidation products, malonaldehyde and diunsaturated aldehyde⁽⁷⁾. The use of naturally occurring food antioxidants has advantages in preventing WOF since they may already present in food. Ramsey and Watts⁽⁸⁾ showed that the extracts of several vegetables act as antioxidants when applied to slices of cooked beef. Although beef stew contains many of the vegetables that act as antioxidants in lipid oxidation, no reports were found of investigations in which the degradation of lipids either by hydrolysis or oxidation was determined in beef stew. Perter-

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son *et al.*⁽⁹⁾ investigated the aroma of canned beef stew and Peterson and Chang⁽¹⁰⁾ compared flavor compounds in fresh, frozen beef stew with those present in canned beef stew. However, neither group of researchers determined the effect of processing and storage on the lipid or fatty acids of beef in the stew. The objective of this study was to assess process and storage effects on beef lipids and fatty acids of polar lipids in beef stew.

Materials and Methods

Experimental plan

Beef stews were prepared by 2 different processes. Stews in process 1 were cooked and put into a closed polyethylene container. Stews in process 2 were cooked and packaged under vacuum in a barrier bag. Stews in each process were prepared by 4 different treatments; beef cooked alone (T1), with onions (T2), with carrots (T3), and with onions and carrots (T4). The storage periods for process 1 and 2 were established by commercial storage conditions for conventional manner and barrier bag. Stews in process 1 were stored for 0, 2 or 4 days at 5°C and stews in process 2 were stored for 0, 2 or 4 weeks at 0°C. Two replications were run for each sample.

Materials

The biceps femoris (BF) and semitendinosus (ST) muscles from the left and right sides of maturity A steer carcasses from a local meat packaging plant were obtained. The muscles were trimmed of outside fat and connective tissue and cut in 1.25 cm cubes. Prior to the start of the experiment, the 500-g packages of beef in each replication were divided at random equally between process 1 and 2 and stored at -35°C. Frozen, sliced carrot (Consolid, D MDT Inc., Atlanta, GA) and (or) freeze dried onions (Basic American Foods, San Francisco, CA) were purchased from Institutional Jobbers, a wholesale institutional food supplier in Knoxville, TN.

Preparation, storage and analysis of stews

The beef stew formulation shown in Table 1 was used as guide for preparation of the beef stews⁽¹¹⁾. However, only those vegetables included for each treatment were used in this study, and flour or starch was omitted from all stews. Prior to the

Table 1. Formula of beef stew from Shinn and Jackson (1979)

Ingredient	Amount
Beef-raw	400 g
Potatoes	300 g
Carrots	200 g
Peas	60 g
Onions	24 g
Salt	16 g
White pepper	2 g
Flour or Starch	36 g
Water or Stock	559 g

cooking of each stew, a 500-g package of beef was thawed for 16 hours at 5°C. One hundred grams of the thawed meat were removed, refrozen, powdered and stored at -18°C until analyzed. Four hundred grams of thawed beef was cooked for beef stew as described by Peterson and Chang⁽¹⁰⁾. For the analysis of beef stew, the beef was separated from vegetables and broth, and vegetables and broth were homogenized. Two hundred grams of the cooked beef were powdered and stored at -18°C until analyzed for all measurements except TBA analysis. Forty grams of the cooked beef were homogenized with eighty grams of homogenized vegetables and broth. This homogenate was analyzed for TBA value.

Total lipid extraction

The total lipids were extracted from 40-g of the raw beef used for each stew and 40-g of cooked beef of each stew by the method of Melton *et al.*⁽¹²⁾.

Phospholipid analysis

The concentration of total lipids in grams per ml of the concentrated lipid extract was determined in duplicate. Then one ml of the concentrated lipid extract was diluted to 4 ml with CHCl₃ and filtered through a 0.45 µl filter. Five µl of this diluted sample were analyzed for levels of the individual phospholipids, cephalin and lecithin. For phospholipid analysis, the method of Jungalwala *et al.*⁽¹³⁾ was modified. This analysis was done on a Waters' High Performance Liquid Chromatograph equipped with a Model 480 UV detector and a Shimadzu data processor using a Whatman Partisil 10 column (length: 270 mm, o.d.: 6.35 mm, i.d.: 4.60 mm) and an acetonitrile: methanol: water (70:20:10, V:V:V) solvent

Table 2. Mean contents of phospholipids of cooked beef in stew for process and treatment

Variable	g/100g Dry beef	
	Cephalin	Lecithin
Process		
1	0.38	1.91 ^{a)}
2	0.39	1.78 ^{b)}
Treatment ^{c)}		
1	0.36	1.75
2	0.35	1.69
3	0.41	1.94
4	0.42	2.00

^{a),b)}For any one variable, means in a column followed by different superscripts are significantly different at the $p < 0.05$ level.

^{c)}For cephalin content, mean separation by orthogonal comparisons resulted in a significant difference between treatments 2 and 3.

containing 0.1% H_3PO_4 at a flow rate of 2.0 ml/min. The contents of phospholipids were measured at 205 nm. A standard curve (peak area versus μg lipid injected) was determined for each of the phospholipid using individual lipid standards prepared from bovine liver (Sigma Chemical Co., Inc., St. Louis, MO).

Fatty acid analysis of phospholipid

Phospholipids were separated from triglycerides by the method of Melton⁽¹⁴⁾. The dried phospholipids were converted to methyl esters by the boron trifluoride method⁽¹⁵⁾. The dried pentane solution containing the methyl esters was analyzed by a gas chromatographic (GC) method using a 30-meter fused silica SP-2350 capillary column (Supelco Inc., Bellefonte, PA). The GC run was temperature programmed from 150 to 220 °C at 2 °C/min on a Shimadzu Model 6 AM gas chromatograph equipped with flame ionization detectors and a Shimadzu E-1B data processor. Selected fatty acid methyl ester samples were also analyzed on the same capillary column using a Shimadzu Model 9 AM gas chromatograph interfaced with a Shimadzu QP-1000 mass spectrometer (MS) set at 70 eV. The temperature program for the GC-MS run was 150 to 188 °C at 2 °C/min followed by a rate of 5 °C/min from 188 to 220 °C.

TBA test

The modified distillate method⁽⁶⁾ was used for

Table 3. Mean phospholipid contents of raw versus cooked beef from stews^{a)}

Phospholipid	g/100g Dry beef	
	Raw(n =16)	Cooked(n = 16) ^{b)}
Cephalin	0.62 ^{b)}	0.38 ^{c)}
Lecithin	2.24 ^{b)}	1.77 ^{c)}

^{a)}Means in a raw followed by different superscripts are significantly different at the $p < 0.05$ level.

^{b)}From stews that were not stored (0-day or 0-week).

determination of lipid oxidation.

Results and Discussion

Phospholipid content

Concentrations of phospholipids (cephalin and lecithin) of cooked beef for each process and treatment are shown in Table 2. Cooked beef from stews in process 1 (conventional) contained higher levels of lecithin than that from stews in process 2 (barrier bag). Orthogonal comparisons for treatments showed that the beef from stews in treatment 2 (beef cooked with onions) had significantly less cephalin content than beef from stews in treatment 3 (beef cooked with carrots). Addition of carrots to the stew offered some protection against loss of cephalin during cooking. Storage did not significantly affect the content of phospholipid.

The mean contents of cephalin and lecithin in raw versus cooked beef are given in Table 3. Cooking the beef resulted in a 39% loss of cephalin and a 21% loss of lecithin, when both were expressed on a dry beef. The loss of these phospholipids most likely was due to hydrolysis since the major degradation products of the phospholipids in the present study were lysocephalin and lysolecithin. Igene and Pearson⁽³⁾ reported that cooking caused significant decreases in total phospholipid of beef, with cephalin being the most labile phospholipid. They indicated that decreases in the levels of phospholipids (mainly lecithin and cephalin) of beef during cooking and storage may be due to either autoxidation, hydrolytic decomposition, lipid browning reactions or lipid-protein co-polymerization.

Fatty acid contents of phospholipid

Mean fatty acid contents of phospholipids of cooked beef for each process are given in Table 4. No significant differences were found among treat-

Table 4. Mean fatty acid contents of phospholipids of cooked beef for each process^{a)}

Fatty acid	mg/100g Dry beef	
	Process 1 ^{b)}	Process 2 ^{c)}
14:0	3.9	3.3
15:0	4.3	4.6
16:0	206.4 ^c	189.0 ^d
16:1	12.2	11.4
117:0	3.2	3.1
17:0	5.5	4.9
17:1	7.1	6.9
18:0	133.1	117.7
118:1	4.5	4.4
18:1	204.3	182.4
118:1	31.4	28.3
18:2	270.3	243.5
18:3	8.8	8.0
20:0	1.0	1.0
20:2	2.7	3.0
20:3	38.7	35.4
20:4w6	132.9	119.1
20:4w3	1.1	1.5
20:5w3	15.3	14.6
22:4	... ^{d)}	... ^{d)}
22:5w6	1.9	1.7
22:5w3	28.6	25.4
22:6w3	3.2	3.2
unknown	28.7	29.6

^{a)}Means in a row followed by different superscripts are significantly different at the $p < 0.05$ level.

^{b)}Conventional way.

^{c)}Barrier bag.

^{d)}Wasn't present.

ments for any fatty acid, and significant storage effect were found for 18:0, 20:2 and 22:5w6 only. The only significant difference in fatty acid content found between processes was for 16:0. The major polyunsaturated fatty acids in this present study were 18:2, 20:4w6, 20:3, 20:5w3 and 22:5w3.

The phospholipid fatty acid contents of raw versus cooked beef in stews are given in Table 5. Most fatty acids were decreased in concentration by cooking except for 18:0 and 22:5w3. Cooking decreased the concentrations of 15:0, 16:1, 17:0, 18:3 and the unknown fatty acids by 64, 25, 76, 21 and 54%, respectively. Keller and Kinsella⁽⁴⁾ reported that the 20:4 fatty acid in cephalin was decreased by 25% during cooking. Also, Love and Pearson⁽¹⁶⁾ reported that the decrease in the 20:4 fatty acid they found was consistent with its greater propensity to undergo autoxidation, especially if it were

Table 5. Mean fatty acid contents of phospholipids of raw versus cooked beef^{a)}

Fatty acid	mg/100g Dry beef	
	Raw (n = 16)	Cooked ^{b)} (n = 16)
14:0	0.9	3.5
15:0	14.5 ^b	5.2 ^c
16:0	203.0 ^b	191.0 ^c
16:1	15.8 ^b	11.8 ^c
117:0	3.0 ^b	2.9 ^c
17:0	21.0 ^b	5.1 ^c
17:1	9.4 ^b	6.8 ^c
18:0	119.6 ^b	122.7 ^c
118:1	2.5	4.4
18:1	189.1 ^b	183.0 ^c
118:1	34.6 ^b	29.4 ^c
18:2	261.7 ^b	250.7 ^c
18:3	10.6 ^b	8.4 ^c
20:0	... ^{b,c)}	1.5 ^c
20:2	1.6	3.1
20:3	40.8 ^b	36.4 ^c
20:4w6	135.4 ^b	124.0 ^c
20:4w3	2.2 ^b	1.4 ^c
20:5w6	18.4	15.3
22:4	1.3 ^b	... ^{c)}
22:5w6	0.8	2.2
22:5w3	25.9 ^b	27.3 ^c
22:6w3	3.0	3.3
unknown	70.0 ^b	28.0 ^c

^{a)}Means in a row followed by different superscripts are different at the $p < 0.05$ level.

^{b)}From stews that were not stored (0-day or 0-week).

^{c)}Wasn't present.

associated with cephalin.

The content of 18:0, 20:2 and 22:5w6 as a function of storage for each process are given in Table 6. Although these acids were significantly affected by storage, there was not a consistent trend in the effect of storage on the concentration of any of these acids.

Fatty aldehyde contents of phospholipid

The concentration of fatty aldehydes were not affected ($p < 0.05$) by process, treatment or storage (Table 7). The aldehydes were tetradecanal (14:0A), pentadecanal (15:0A), hexadecanal (16:0A), hexadecenal (16:1A), octadecanal (18:0A) and octadecenal (18:1A). The most abundant fatty aldehyde in beef phospholipids was 16:0A, followed by 18:0A. The levels of all aldehyde, except 15:0A and 18:0A, were decreased by cooking (Table 8). The loss in concentration of these fatty aldehydes was most

Table 6. Mean concentrations of some of beef phospholipid fatty acids for each process during storage

Fatty acid	Process	mg/100g Dry beef		
		Storage ^{a)}		
		0	2	4
18:0 ^{b)}	1	136.0	124.4	139.1
	2	109.4	114.1	129.7
20:2 ^{b)}	1	3.1	1.7	3.3
	2	3.1	2.8	3.1
22:5w6 ^{b)}	1	2.2	1.4	2.3
	2	2.2	1.4	1.6

^{a)}Process 1 was stored for 0 to 4 days at 5 °C and process 2 was stored for 0 to 4 weeks at 0 °C.

^{b)}The contents were affected by storage ($p < 0.05$).

Table 7. Mean concentrations of phospholipid fatty aldehydes in cooked beef for each process

Aldehyde	mg/100g Dry beef	
	Process 1 ^{a)}	Process 2 ^{b)}
14:0A	4.0	2.9
15:0A	2.4	2.4
16:0A	76.2	76.3
16:1A	5.0	5.2
18:0A	45.0	44.3
18:1A	8.8	9.0

^{a)}Conventional way.

^{b)}Barrier bag.

likely due to hydrolysis during cooking. The increase in levels of 15:0A and 18:0A by cooking is hard to explain; however, it could be due to non-homogeneity in the beef sample.

Malonaldehyde content

The mean malonaldehyde (MA) contents of cooked beef stew for each process during storage are shown in Table 9. The MA content in process 1 stews was affected ($p < 0.05$) by storage according to the following equation $Y = 1.82 + 0.2431X$ ($Y =$ MA, $X =$ days) while process 2 was not affected ($p < 0.05$) by storage. The mean MA content of raw beef in the present study was 0.79 mg/kg beef. This value is higher than that of fresh beef and is probably due to the fact that the beef was stored at -18 °C for approximately 3 to 4 months prior to the start of the experiment. Comparison of the MA content of raw beef with that of the cooked stew shows that cooking increased MA content greatly. These results are in agreement with many other investiga-

Table 8. Mean concentrations of phospholipid fatty aldehydes of raw versus cooked beef^{a)}

Aldehyde	mg/100g Dry beef	
	Raw (n = 16)	Cooked ^{b)} (n = 16)
14:0A	16.8 ^{b)}	4.3 ^{c)}
15:0A	1.5 ^{b)}	2.2 ^{c)}
16:0A	90.1 ^{b)}	79.0 ^{c)}
16:1A	6.9 ^{b)}	5.6 ^{c)}
18:0A	43.2 ^{b)}	46.3 ^{c)}
18:1A	10.5 ^{b)}	9.4 ^{c)}

^{a)}Means in a row followed by different superscripts are significantly different at the $p < 0.05$ level.

^{b)}From stews that were not stored (0-day or 0-week).

Table 9. Mean malonaldehyde contents of cooked beef stew as affected by process and storage

Process	Storage ^{a)}		
	0	2	4
	mg MA/kg stew, wet basis		
1 ^{b)}	1.67	2.58	2.65
2 ^{c)}	1.36	1.86	1.98

^{a)}Process 1 was stored for 0 to 4 days at 5 °C and Process 2 was stored for 0 to 4 weeks at 0 °C.

^{b)}Significantly affected by storage according to the following equation $Y = 1.82 + 0.2431X$ where $Y =$ malonaldehyde content and $X =$ days ($p < 0.05$).

^{c)}Not significantly affected by storage ($p < 0.05$).

tors^(1,3,7). Malonaldehyde is a secondary oxidation product of polyunsaturated fatty acids and its concentration is expressed as the TBA value, mg of MA per kg of sample⁽⁷⁾. The TBA value is the most popular measurement of lipid oxidation in muscle foods⁽⁷⁾ and is an effective measurement of oxidation during storage and cooking of meat. The results of the present study indicate that the barrier bag at 0 °C was superior to the polyethylene container at 5 °C for prevention of lipid oxidation in beef stews during storage. However, although the TBA value indicated that oxidation occurred during storage of process 1 stews, accompanying decreases in levels of polyunsaturated fatty acid of the phospholipids during storage were not found.

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소고기 Stew에 야채첨가가 지방분해에 미치는 영향

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소고기 Stew의 Process, treatment과 storage가 Stew의 지방분해에 미치는 영향을 조사했다. Process는 Stew을 조리한 후 Polyethylene 용기에서 5°C에서 저장한 것(P1)과 barrier에 0°C에서 저장한 것(P2)을 이용했으며, treatment은 Stew을 만들 때 소고기(T1), 소고기와 양파(T2), 소고기와 당근(T3) 그리고 소고기, 양파 및 당근(T4)을 넣어 만들었다. P1 Stew는 0, 2, 4일간 저장했으며, P2 Stew는 0, 2, 4주 동안 저장하였다. 조리한 소고기 Stew는 조리하지 않는 것에 비교해서 ce-

phalin 양이 39%, lecithin 양이 21% 감소했으며, 그외 대부분의 인지질과 지방산도 감소했다. Process의 차이나 storage의 차이에 따라 cephalin이나 lecithin의 양에 현저한 영향을 미치지 않는으나 소고기와 당근으로 만든 Stew에서 당근이 cephalin의 가수분해를 약간 억제했다. 특히, P1 Stew가 P2 Stew보다 높은 TBA가를 나타냈으며, P1 Stew의 TBA가는 4일간 저장하는 과정에서 계속해서 증가했으며, P2 Stew의 TBA가는 4주 동안 현저한 변화가 없었다.