Effect of Castration on Biochemistry and Quality of Beef

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Under comparable feeding management involving good nutrition from birth, bulls are more efficient in converting feed to meat than steers. Furthermore, bulls produce a higher percentage of edible meat than steers, the difference being due primarily to the waste fat from steers rather than other body characteristics. Carcass grade and tenderness of the meat of bulls were slightly less. However, differences in tenderness and general consumer quality were perhaps not commensurate with increased cost of producing meat from steers for most consumers. The use of diethylstilbestrol on bulls improved the efficiency of meat production still more and the carcass grades and the consumer quality of the meat approached that for the steers. The relationship of color (cyanometmyoglobin), marbling (intramuscular fat), connective tissue (hydroxyproline), and muscle plasma proteins to the major consumer quality attribute of tenderness and to carcass grade has been studied. Muscle plasma proteins greatly affect tenderness and this is even more pronounced when connective tissue is low, as in young well fed animals. Marbling has no relation to tenderness, except in so far as it may indicate sex, breeding, quality of the ration, and length of feeding period. Color has no relation to tenderness, except that it may reflect some age or sex differences.

HORMONES are well known among biochemists, physiologists, and nutritionists to have a major role in rate of growth and efficiency of feed utilization. They also play a part in the quality of meat produced by an animal. The widely practiced manifestation of these principles is the practice of castrating meat-producing animals, particularly males. However, the relation of castration to the efficiency of meat production and the quality of meat produced has not been critically investigated. Such information is important in view of the rapid rate of growth of our population, present meat production practices, and the American taste for good meat. To study this problem effectively it is necessary to study meat-producing animals all the way through breeding, feeding, production management, slaughtering, and cutting into consumer units. Furthermore, consumer quality of the resulting meat should be investigated. This kind of study requires an integrated effort by many interested in the ultimate purpose of producing more and better meat cheaper, and serves both farmer (producer) and consumer. It is also appropriate for land grant colleges and experiment stations.

The work described here is only part of a continuing program on the effect of castration and includes for the most part only information on the yield, quality, and biochemistry of meat from steers, bulls, and diethylstilbestrol-treated bulls bred and fed under comparable conditions. Details of breeding and feeding management will appear elsewhere (*11*). The data were obtained over a 3-year period. In each year Hereford bull calves were selected from western herds of rather uniform breeding. They were born in late March and early April, carried in their home herds until autumn, and fed at The Ohio Agricultural Experiment Station at Wooster on high quality rations until they weighed approximately 900 pounds. The animals were then slaughtered at 13 to 15 months of age. The slaughtering and carcass studies were conducted at Columbus.

Methods

Most of the methods used to obtain data have been noted in other publications from this laboratory (*2, 7, 9, 10, 16*).

"Edible portion" as used here refers to meat usually consumed and is determined in accordance with procedures given by Hershberger and coworkers (*7*). The meat was separated from bone and waste fat, so that the lean meat carried no more fat covering than $^1/_4$ to $^3/_8$ inch. The waste fat is that usually trimmed off during retailing, plus that which usually reaches the garbage can of the consumer.

All chemical data were obtained from the longissimus dorsi in the same relative position as the meat taken for taste testing for tenderness. The following methods were used for determining intramuscular fat and color of meat as represented by hemoglobin and myoglobin.

Fat, Intramuscular. The meat grader and consumer attach much importance to marbling. From the chemist's standpoint this is represented by intramuscular fat determinations, although such data do not tell the distribution of fat within the muscle. A rapid method for determining fat in fresh and processed meats was developed by Hoover (*8*) in this laboratory, based essentially on the work of Glimm and Bauer (*6*) with the exclusion of butanol.

Approximately 5 grams of ground meat sample were weighed into a 9-gram Paley 20% cream test bottle. To the meat was added 30 ml. of the alkaline digesting solution, prepared as needed from the following solutions.

A. 240 grams of sodium potassium tartrate in 1000 ml. of water
B. 90% ethyl alcohol
C. 200 grams of sodium hydroxide in 1000 ml. of water.

To give the protein digesting solution 440 ml. of A, 264 ml. of B, 210 ml. of C, and 176 ml. of water were mixed. The bottle and contents were heated in a water bath at 75° to 80° C. until the meat particles dissolved. Additional alkaline digesting solution was poured into the neck of the bottle and then centrifuged for 3 minutes. Hot water was added to bring the surface of the fat

layer to within 1 to 2 cm. of the top of the neck. This was followed by 2 more minutes of centrifugation. The fat column was read after 2 minutes' warming in a 70° C. water bath. Readings were made from the top of the upper meniscus to the bottom of the lower meniscus and the fat content of meat was computed in accordance with sample size and bottle calibration. Results agree well with the usually accepted ether extraction methods and much time is saved by this procedure.

Cyanometmyoglobin. Myoglobin is quantitatively the most important pigment in meat, although hemoglobin is present in amounts of 10% more or less of the myoglobin content of meat (*13*, *15*). Because of the press of time in trying to get as much information as possible on fresh samples, myoglobin plus hemoglobin was determined. Consequently, the data given in this paper for cyanometmyoglobin are for cyanometmyoglobin plus cyanomethemoglobin and, therefore, should be adjusted downward for myoglobin only. However, the data as they are serve well for comparative purposes. Only slight modifications from previously published methods used in the laboratory (*7*, *9*) were made, as suggested by Drabkin (*4*).

A 10-gram meat sample was mixed for 3 minutes with 90 ml. of water in a Waring blender. This was filtered through quantitative paper and the first few drops, which were cloudy, were discarded. To the filtrate 50 mg. of potassium ferricyanide were added and dissolved; then, similarly, 25 mg. of potassium cyanide. By this procedure the pigments were converted to the cyanometmyoglobin and cyanomethemoglobin. Next 5 mg. of calcium chloride hexahydrate were added to flocculate any colloidal dispersion. After 5 minutes the solution was filtered and the absorbance was determined using a Beckman DU spectrophotometer with a 10-mm. cell. Based on the spectral characteristics of Drabkin (*4*) and the dilution factors, it can be shown that total pigment on the wet basis as cyanometmyoglobin is given by absorbance at 540 mμ multiplied by 1.456.

Animals Studied

Experiment 1, 1950–1951. As soon as the 30 experimental animals were chosen, 10 were castrated on May 11, 1950. When the animals reached Wooster, 10 more were castrated, leaving 10 as bulls. As the animals reached market weight two were chosen from each group and slaughtered at weekly intervals. The bulls gained weight more rapidly than the steers and consequently were significantly heavier.

Experiment 2, 1951–1952. This experiment duplicated in part the 1950–51 experiment. Thirty-six animals were used. Twelve were castrated on March 21 or April 19, 1951, 12 were castrated on November 7, 1951, and 12 remained as bulls. The slaughtering schedule was originally set up to use the animals as they reached 900 pounds. The bulls gained faster and were slaughtered earlier. Because of the hot summer weather the steers ate and gained less rapidly and a large share of them did not reach the desired weight. It was decided that the differences in age would be a dominant factor if the steers were carried to the desired weight, and, therefore, the experiment was terminated with steers significantly lighter in weight though somewhat older.

Experiment 3, 1952–1953. Forty-eight bull calves were used. This year several variations of treatment were introduced: Early castration was discontinued; a few were castrated rather late, at 10 months of age; diethylstilbestrol implantation in the ear was used in place of castration; and the bulls were divided into light and regular weight to determine whether the lightweight bull calves would have characteristics closer to steers in terms of cutout, hide, and head weights. As this did not prove to be so, the data for all 20 are pooled for part of the data concerned with cutout value. The 48 animals were divided as follows: Eight were castrated October 16, 1952; 5 were treated on October 22, 1952, and on January 29, 1953, with 25 mg. of diethylstilbestrol per cwt.; 10 were implanted on January 29, 1953, only; 5 were castrated on January 29, 1952; and 20 remained as bulls. The bulls were separated into two groups—10 were slaughtered at lightweight (approximately 800 pounds) and 10 were slaughtered at regular weight. The slaughtering schedule was on a weekly interval of six per week in so far as possible.

Table I. Effect of Castration and Hormone Treatment on Daily Rate of Gain, Carcass Grade, Yield of Edible Meat, Bone, Hide, and Waste Fat, and Tenderness of Meat

	1950–1951			1951–1952			1952–1953				
	Castrated			Castrated			Castrated			Diethylstilbestrol	
	5-11-50	10-31-50	Bulls	3-21, 4-19-51	11-7-51	Bulls	10-16-52	1-29-53	Bulls	1-29-53	10-22-52, 1-29-53
No. of animals	10	10	10	12	12	12	8	5	20	10	5
Av. daily gain, lb.	2.00[a]	2.00[a]	2.23[a]	1.96[b]	1.94[b]	2.43[c]	2.07[d]	1.84[e]	2.36[f]	2.63[e]	2.69[d]
Av. live wt., lb.	867	859	929	817	823	848	847	873	856[g]	958	867
Av. chilled carcass[h], lb.	529	526	556	489	498	516	504	509	501	574	515
Dressing percentage[i]	61.0	61.2	59.8	59.9	60.3	60.7	59.5	58.2	58.5	60.0	59.3
Fore quarter, %[j]	52.2	51.5	53.8	51.5	51.5	53.4	51.0	51.0	52.8	53.4	52.0
Hind quarter, %[j]	47.8	48.5	46.2	48.5	48.5	46.6	49.0	49.0	47.2	46.2	48.0
Edible portion, %[j]	73.7	74.1	77.7	74.1	74.4	77.5	73.2	73.9	77.5	77.3	75.8
Edible portion, %[k]	45.0	45.3	46.5	44.4	44.9	47.0	43.5	43.0	45.3	46.4	45.2
Waste fat, %[j]	10.4	10.0	5.5	9.8	9.4	5.7	11.3	9.9	6.3	7.7	8.2
Bone, %[j]	15.4	15.6	16.2	16.1	16.2	16.4	15.5	16.2	16.1	15.0	15.8
Head, %[k]	3.1	3.1	3.2	3.1	3.1	3.2	3.3	3.4	3.4	3.2	3.1
Hide, %[k]	8.0	8.4	9.5	8.3	8.1	9.6	8.2	8.9	9.6	8.5	8.5
Carcass grade[l]	0.80	1.03	2.23	1.43	1.44	2.78	1.33	1.82	2.24	2.09	1.64
Tenderness, 15 days	8.11	7.62	7.57	7.62	7.74	6.89	8.01	7.58	7.27	6.81	7.72

[a] 252 days' feeding test.
[b] 247 days' feeding test.
[c] 210 days' feeding test.
[d] 196 days' feeding test.
[e] 98 days' feeding test.
[f] 140 and 196 days' feeding tests pooled.
[g] 10 lightweight 801 lb.
[h] Hot carcass, 2.5%.
[i] Chilled carcass/live wt.
[j] Based on carcass wt.
[k] Based on live wt.
[l] Numerical U. S. grade factors: 0.0 top prime, 0.4 prime, 0.7 low prime; 1.0 top choice, 1.4 choice, 1.7 low choice; 2.0 top good, 2.4 good, 2.7 low good; 3.0 top comm., 3.4 comm., 3.7 low comm.

Table II. Tenderness, Carcass Grade, and Biochemical Characteristics of Steers, Bulls, and Bulls Treated with Diethylstilbestrol

	1950–1951			1951 1952			1952–1953					
	Steers Castrated			Steers Castrated			Steers Castrated		Bulls		Stilbestrol Implanted	
	5-11-50	10-31-50	Bulls	3-21, 4-19-51	11-7-51	Bulls	10-16-52	1-29-52	Light wt.[a]	Reg. wt.[b]	10-22-52 1-29-53	1-29-53
No. of animals	10	10	10	12	12	12	8	5	10	10	10	5
Carcass grade	0.80	1.03	2.33	1.43	1.44	2.78	1.33	1.82	2.47	2.01	2.09	1.64
St. deviation	0.38	0.38	0.26	0.64	0.56	0.19	0.30	0.16	0.59	0.43	0.32	0.63
St. error	0.12	0.12	0.08	0.20	0.18	0.05	0.10	0.07	0.18	0.14	0.10	0.28
Tenderness, 3 days	7.29	6.52	5.58	6.53	6.42	5.11	5.77[c]	...	...	...	5.28[d]	...
St. deviation	0.78	0.59	0.90	0.96	0.55	0.78	0.74	...	...	...	1.03	...
St. error	0.25	0.19	0.29	0.27	0.16	0.23	0.43	...	...	...	0.52	...
Tenderness, 15 days	8.11	7.62	7.57	7.62	7.74	6.89	8.00	7.58	7.09	7.45	6.77	7.72
St. deviation	0.49	0.87	0.66	0.29	0.67	0.75	0.64	0.39	0.69	0.65	0.62	0.94
St. error	0.16	0.28	0.21	0.08	0.19	0.22	0.23	0.17	0.22	0.12	0.20	0.42
Fat, intramuscular, %	6.08	6.64	2.88	5.18	4.46	2.30	3.76	2.33	1.91	2.23	2.35	1.93
St. deviation	1.27	1.30	0.77	0.98	1.65	0.63	1.83	0.73	0.56	0.62	0.59	0.68
St. error	0.40	0.41	0.24	0.28	0.48	0.18	0.63	0.33	0.18	0.20	0.18	0.30
Cyanometmyoglobin, %[e]	0.432	0.409	0.470	0.439	0.461	0.389	0.363	0.343	0.332	0.378	0.351	0.276
St. deviation	0.081	0.078	0.071	0.067	0.073	0.037	0.040	0.053	0.047	0.063	0.046	0.031
St. error	0.026	0.029	0.023	0.020	0.021	0.011	0.014	0.024	0.015	0.020	0.014	0.014
Hydroxyproline, %	0.0445	0.0507	0.0546	0.0503	0.0498	0.0532	0.0379	0.0273	0.0301	0.0438	0.0357	0.0215
St. deviation	0.0051	0.0048	0.0069	0.0115	0.0078	0.0074	0.0067	0.0032	0.0053	0.0086	0.0069	0.0027
St. error	0.0016	0.0016	0.0022	0.0034	0.0023	0.0021	0.0022	0.0015	0.0017	0.0027	0.0022	0.0012
Total nitrogen, %	...	...	...	...	...	...	3.348	3.400	3.595	3.383	3.416	3.382
St. deviation	...	...	...	...	...	...	0.123	0.129	0.150	0.099	0.104	0.065
St. error	...	...	...	...	...	...	0.044	0.058	0.048	0.032	0.033	0.030
% Total N extracted	...	...	...	...	...	...	29.20	27.22	23.98	25.29	25.25	29.04
St. deviation	...	...	...	...	...	...	4.15	0.72	3.44	3.55	1.54	3.19
St. error	...	...	...	...	...	...	1.47	0.32	1.09	1.12	0.48	0.41
pH, 3 days	5.52	5.58	5.59	5.59	5.61	5.47	...	...	...	...	...	...
St. deviation	0.35	0.13	0.10	0.07	0.05	0.07	...	...	...	...	...	...
St. error	0.11	0.04	0.03	0.02	0.02	0.02	...	...	...	...	...	...
pH, 15 days	...	...	...	5.67	5.70	5.73	5.63	5.48	5.58	5.60	5.54	5.38
St. deviation	...	...	...	0.03	0.03	0.04	0.05	0.05	0.09	0.07	0.10	0.03
St. error	...	...	...	0.01	0.01	0.01	0.02	0.02	0.03	0.02	0.03	0.02

[a] Average live weight 801 lb.
[b] Average live weight 911 lb.
[c] Four animals only.
[d] Five animals only.
[e] Values represent both hemoglobin and myoglobin.

Results and Discussion

Carcass Characteristics and Growth Data. Table I summarizes the pertinent data on carcass and rates of gain. The economics of meat production rest in the cost of producing a particular carcass (*11*). However, for the purposes at hand attention is called to the average daily gain. As all animals were on comparable rations, these data indicate relative production costs—the higher the rate of daily gain the lower the cost of production. It is at once apparent that the bulls were more efficient gainers than steers and that hormone-treated bulls were the most efficient of all.

Under livestock marketing practices bulls usually bring less than steers. Farmers' profits are determined by the difference between production costs and selling price. Even though the bulls brought less on the market, they might still be more profitable than the steers. In a year when cattle are scarce, such as 1950–1951, the differential in profits for bulls is greater than in a plentiful year such as 1952–1953.

The dressing percentage is usually considered higher for steers than bulls. This was true only for the 1950–51 series, when animals were killed at the same age. It was not true in the other years, when tests were terminated on the basis of animal size.

The yield of edible portion was significantly greater each year for the bulls than for the steers, whether expressed on the live animal or carcass basis. On the carcass basis bulls yielded 3 to 4% more meat than steers. In bulls there is less waste fat and this more than offsets the slightly higher dressing percentage for steers. Furthermore, the greater yield of meat from bulls more than outweighs the slightly greater amount of bone, head, and hide. From the packer's point of view heavier hides, such as those produced by bulls, bring less at the present time than light ones on a per pound basis.

It is usually considered that bulls have less of the desirable hind quarter than steers. Yet, if the fact that the kidney knobs of bulls are much smaller than those of steers (not shown in Table I) is considered, there is essentially no difference between hind quarter yield of bulls and steers. This means that the bulls yielded the same amount of steaks and rounds as the steers.

The U. S. carcass grades were about 1.3 points higher for steers than for bulls for the first two years, but this difference was less in the third year. In fact, the bulls doubly implanted with diethylstilbestrol gave carcasses grading the same as the steers. Many factors go into grading carcasses, but the grade standards were established on steers and bulls are usually downgraded because they are bulls and not merely because they carry less fat.

The eating quality as seen in the tenderness rating (the primary attribute of quality for the consumer) indicates that the bulls yielded slightly less tender meat. Yet the differences observed were rather small though significant. There was no doubt that the bulls yielded very desirable meat. Perhaps the least desirable meat of the 3-year study was that from the singly implanted bulls, 1952–1953. The hormone had been used up well before slaughter time and these animals were more "bully" than the other young

bulls and gave the appearance of being older than they actually were. To counterbalance this adverse effect of insufficient hormone, once such treatment was started, was the quality of the meat from the doubly implanted bulls, which was in every way as good as the meat from the steers.

Biochemical Characteristics and Tenderness. This study on effect of castration offered an unusual opportunity to study the biochemistry of meat in relation to eating quality, a project that is under active investigation in this laboratory. Table II shows most of the biochemical data obtained. Inasmuch as the detailed data on all 114 animals are available, the authors give the mean values for each group on a particular treatment together with the standard deviation and standard error for each mean. This gives some idea of the spread in individual values and a basis for determining significances of differences.

Tenderness was determined at both 3 and 15 days post mortem for the first 2 years and at 15 days only for most of the animals the third year. Most of the improvement in tenderness due to aging occurs within the first 17 days' storage at 33° F. (*7*). Consequently, 15 days' storage at 37–38° F. was used in this work, as it fitted well into the general plan of the experiment. As the tenderness at 3 to 4 days post mortem for the first 2 years' study agreed very well, the press of securing other data in the third year made it advisable to drop the taste testing at 3 days. Even though most consumer beef is not intentionally aged, it is usually 7 to 14 days post mortem in normal distribution before it is consumed, and therefore, tenderness determination at 15 days' aging at 37–38° F. was considered more analogous to normal practice.

Bulls produced only slightly less tender meat than steers and this difference was found at both 3 and 15 days. Even at 3 days the meat was not objectionably tough. As all animals were young and well fed, the tenderness data simply confirm the usually accepted idea that in producing tender meat, age of animal and quality of ration are perhaps the most important items in feeding management. In other words, animals fed well from birth and marketed young produce tender meat. Even so, this does not tell what is tender meat in fundamental biochemical and physiological terms.

Marbling has long been considered a mark of quality in meat. Data on intramuscular fat show very clearly that steers carry more than bulls for the most part, although the late castrates of the third year were low and comparable to the bulls. This may be a reflection of the late date for castration and more pronounced shock effect of the operation. The effect of the diethylstilbestrol is noteworthy, in that the single implants were comparable to the bulls in intramuscular fat and the doubly treated bulls had a very low fat content, notwithstanding the high tenderness of the meat and carcass grade comparable to the steers. Whether this low fat is a specific hormone effect or due simply to increased anabolism is not known.

Color of meat is an important factor in consumer appeal, and an earlier report (*9*) indicated that color might be an index to total muscle substance or improvement in tenderness. Furthermore, it was thought that bull beef was more intensely colored than steer beef. This seemed to be borne out by the first year's study, but, just the reverse appeared to be true in the second year. Yet in the first year all animals were killed at approximately the same age and in the second the steers were older than the bulls when slaughtered. Perhaps, then, two factors may contribute to color—age of animal and sex. In the third year the light bulls had less color than the heavier ones. The singly implanted bulls had color comparable to the steers for the third year and the doubly treated bulls were remarkably light in color. Dinusson and coworkers (*3*) observed that diethylstilbestrol depressed erythrocyte count and Wilkinson and coworkers (*18*) have recently shown that diethylstilbestrol-treated sheep have lower hematocrit values. Presumably hemoglobin and perhaps myoglobin formation are depressed by the hormone. Even so these animals were younger when slaughtered, because of their faster gains. It appears that both age and hormones affect meat color and these animals for the most part show less total pigment than those reported previously (*7*, *9*, *15*). Again the present report represents younger animals. When groups of steers or bulls are compared separately, age of animal seems to show closer relation to total color.

Because connective tissue has often been related to tenderness, methods for its chemical determination were sought. Husaini (*9*, *10*) reported on alkali-insoluble protein as an index of connective tissue. This was satisfactory to some extent, but was subject to large error when fat content of the sample was high. Accordingly, hydroxyproline determination was found more satisfactory (*16*). For the first year alkali-insoluble protein and hydroxyproline were compared at 3 and 15 days post mortem. No significant changes took place in either. If there had been enzymic hydrolysis of the connective tissue on aging, a decrease in alkali-insoluble protein should have been found. None was found, and other factors had to be studied with reference to tenderness improvement with post mortem age.

The bulls seemed to have more hydroxyproline than the steers when compared on a yearly basis. For the third year the lightweight bulls had less than the regular weight bulls. The late castrates and light bulls had less than the early castrates and singly implanted bulls. The pronounced effect of the hormone treatment on connective tissue is apparent and the doubly treated animals have a remarkably low hydroxyproline and total connective tissue. All animals in this report were somewhat low in hydroxyproline compared to many market animals of unknown history. No doubt, age and feeding management practices play a part.

In contrast to the varying amounts of hydroxyproline, two amino acids are remarkably constant in meat. As reported earlier (*16*), it was necessary to determine tryptophan and tyrosine as interfering substances for hydroxyproline determinations. On the wet basis the tryptophan content for the groups in the 1950–1951 study were, respectively, in the order of Table II, 0.329, 0.330, and 0.330, with standard deviations of 0.016, 0.013, and 0.019 and standard errors of 0.005, 0.004, and 0.006. Tyrosine contents for the three groups of the 1951–1952 study were in the order of Table II, 1.00, 1.02, and 1.06, respectively, and standard deviations were 0.03, 0.01, and 0.06 with standard errors of 0.01, 0.00, and 0.02. When considered on a nonfat basis the tyrosine values are even more constant between groups.

Table III. Summary of Data for 1950–51

	Total Number	Mean	St. Dev.	St. Error	r^a 3-day	r^a 15-day
Tenderness						
3-day	30	6.45	0.95	0.17	...	...
15-day	30	7.79	0.69	0.13	...	...
Carcass grade	30	1.35	0.71	0.13	−0.667[b]	−0.331[b]
Fat, intramuscular	30	5.17	2.08	0.38	0.138	0.123
Cyanometmyoglobin	30	0.437	0.067	0.012	−0.367	0.083
Hydroxyproline	30	0.0499	0.0069	0.0013	−0.645	−0.575
Alkali-insoluble protein						
3-day	28	0.285	0.129	0.025	−0.526	...
15-day	30	0.312	0.138	0.026	...	−0.223
For 28 degrees of freedom. Probability 0.01, r = 0.463 0.05, r = 0.361						

[a] Correlation coefficient with tenderness.
[b] Negative sign due to inverse order of grade factors.

The facts that no changes were found in connective tissue (alkali-insoluble protein) with post mortem age (*17*) and that classical methods of analysis indicated practically no proteolysis with post mortem age (*9*, *10*) led Wierbicki and coworkers (*17*) to investigate protein fractionation to determine, if possible, the direct relation of muscle plasma to tenderness. The basis for the fractionation was a pH 5.6 buffer designed to extract other plasma proteins and leave actomyosin behind with the thought that the relatively insoluble actomyosin of high molecular weight might be the protein most responsible for toughness. Actomyosin is formed in muscle contraction and actin, myosin, and actomyosin are the proteins reacting in contracting and relaxing muscle (*14*). The actomyosin factor in toughness would be in addition to that connective tissue. In the present study connective tissue was low and apparently contributed relatively little to toughness, thus accentuating the substances in muscle plasma contributing to toughness or its opposite, tenderness. The first attempt at such a study was carried out using the animals from 1952–1953 (*17*); in order to complete the picture here, these data are summarized in Table II. Total nitrogen was determined together with percentage of total nitrogen extracted by the buffer. Total nitrogen is constant for all six groups in 1952–1953, but there is a variation in the amount of nitrogen extracted both within and between groups. The double implants and both groups of steers showed larger amounts extractable than the other groups. It is also apparent when comparing tenderness values that there may be some relation between extractable nitrogen and tenderness. The nature of this relation is being studied and will be reported later.

pH values are what might be expected for meat of the kind studied. However, the pH of the meat from the doubly implanted bulls is significantly lower than the meat from the other animals. The reason for the higher acidity was not determined. As the data on extractability of protein were being collected to determine if a relation existed between actomyosin and tenderness, it was proposed that tenderization with post mortem age might be due to dissociation of some actomyosin to myosin and actin during resolution of rigor mortis. If this were true, the work of Kuschinsky and Turba (*12*) would indicate that there should be an increase in pH with post mortem age. Data bearing on this point were available by chance on the 1951–1952 study and are recorded in Table II. There is a small but significant increase in pH from 3 to 15 days post mortem; not one of the 36 animals showed a decrease in pH over the holding period. Whether this increase in pH is due to dissociation of actomyosin or other factors is not now clear, inasmuch as bacterial factors may be ruled out.

Interaction of Biochemical Factors, Carcass Grade, and Tenderness. In Tables III, IV, and V are summaries of the data for each of 3 years of the study. The possible interactions of biochemical factors and carcass grade with tenderness are indicated as correlation coefficients, *r*. Values of *r* for 1 and 5% probabilities are given.

Carcass grade seems to be a good indication of tenderness at 3 days, as shown for the first 2-year study (Tables III and IV). Grade appears to be less closely related to tenderness at 15 days, although the relation is significant. A bias factor exists to promote a high correlation, in that bulls are usually downgraded.

Marbling or intramuscular fat is commonly thought to be closely related to tenderness. However in the first year's work practically no relation existed between these factors at either 3 or 15 days (Table III). Yet in the second year a very good positive relationship was found at 3 days, which was not maintained at 15 days (Table IV). Table V indicates a fair correlation between intramuscular fat and tenderness at 15 days in the 1952–1953 study. Certainly the relationship is not as close as might be desirable, and again a bias has been introduced in that steers are fatter than bulls at the same age and market weight.

Color as represented by cyanometmyoglobin showed essentially no correlation with tenderness in each of the 3 years, except possibly with the 3-day tenderness in the first year (Table III), but this was not confirmed in the second year (Table IV).

Connective tissue, as hydroxyproline, showed a very good negative correlation with tenderness at both 3 and 15 days in the 1950–1951 study (Table III). A better relation existed than when connective tissue was estimated as alkali-insoluble protein. In view of the high value of the correlation coefficient in the first year, it was something of a contrast to see practically no relation the second year (Table IV) or the third year (Table V). Two factors may contribute to this apparent discrepancy. First, in the 1950–51 study all animals were slaughtered at the same age, whereas in the other years animals were killed as a certain market weight was reached. Secondly, all animals had comparatively low hydroxyproline values and the contribution of this tissue to the tenderness or toughness was not pronounced.

The best relation of any factor with tenderness in the third year of the study was the percentage of extractable nitrogen. This tends to support the view that muscle plasma proteins have an important bearing on the tenderness of meat. The low level of connective tissue (as hydroxyproline) tends to accentuate role of muscle plasma. Although this is the first direct chemical measurement of the role of muscle plasma proteins in tenderness, these results are rather limited and need further confirmation. Furthermore, the nature of tenderness and post mortem changes in tenderness is still unknown. However, a number of avenues of study are at once apparent (*17*).

Table VI gives a summary of the data for the entire study. Although such treatment does not consider all varia-

Table IV. Summary of Data for 1951–52

(Total number of animals, 36)

	Mean	St. Dev.	St. Error	r^a 3-day	r^a 15-day
Tenderness					
3-day	6.02	0.99	0.17	...	...
15-day	7.42	0.72	0.12	...	...
Carcass grade	1.88	0.83	0.16	−0.571[b]	−0.413[a]
Fat, intramuscular	3.98	1.68	0.28	0.590	0.226
Cyanometmyoglobin	0.430	0.067	0.011	0.156	0.249
Hydroxyproline	0.0510	0.0089	0.0015	−0.173	−0.252

For 34 degrees of freedom. Probability 0.01, r = 0.413
0.05, r = 0.321

[a] Correlation coefficient with tenderness.
[b] Negative sign due to inverse order of grade factors.

Table V. Summary of Data for 1952–53

(Total number of animals, 48)

	Mean	St. Dev.	St. Error	r^a, 15-Day
Tenderness, 15-day	7.37	0.76	0.11	
Carcass grade	1.95	0.55	0.08	−0.392[b]
Fat, intramuscular	2.40	1.06	0.17	0.357
Cyanometmyoglobin	0.346	0.054	0.008	0.084
Hydroxyproline	0.0349	0.0080	0.0013	−0.177
Total N extracted, %	26.25	3.55	0.51	0.507

For 46 degrees of freedom. Probability 0.01, r = 0.368
0.05, r = 0.285

[a] Correlation coefficient with tenderness.
[b] Negative sign due to inverse order of grade factors.

Table VI. Summary of Combined Data for 3 Years

Quality Factor	No. of Animals	Mean	St. Dev.	St. Error	r^a, 15-Day	Probability
Tenderness, 15-day						
Entire group	114	7.50	0.74	0.07	...	...
Bulls only	42	7.25	0.72	0.11	...	...
Steers only	57	7.78	0.63	0.08	...	...
Carcass grade						
Entire group	114	1.77	0.73	0.07	−0.425[b]	<0.01
Bulls only	42	2.39	0.48	0.07	−0.297	0.10–0.05
Steers only	57	1.27	0.56	0.07	−0.199	0.2 –0.1
Fat, intramuscular, %						
Entire group	114	3.63	1.94	0.18	0.325	<0.01
Bulls only	42	2.38	0.73	0.11	0.079	0.5
Steers only	57	5.24	2.41	0.30	0.035	0.5
Cyanometmyoglobin, %						
Entire group	114	0.396	0.074	0.007	0.153	0.10–0.05
Bulls only	42	0.392	0.070	0.011	0.244	0.10
Steers only	57	0.418	0.073	0.010	0.085	0.5
Hydroxyproline, %						
Entire group	114	0.0440	0.0110	0.0010	−0.107	0.30–0.20
Bulls only	42	0.0459	0.0121	0.0019	−0.035	−0.5
Steers only	57	0.0457	0.0102	0.0013	−0.043	−0.5

[a] Correlation coefficient with tenderness.
[b] Negative sign due to inverse order of grade factors.

tions in each year's study, some very pertinent generalizations are possible. Furthermore, there are a sufficient number of steers and bulls for a more direct comparison of numbers.

Perhaps the most striking thing is that the young steers were only slightly more tender than bulls of the same age. Recalling (*2*) that a score of 8 is considered "tender" and 6 is only "slightly tough" the half point spread between bulls and steers is rather small even if significant. When one considers the cost of producing steer meat as compared to bull meat on young well fed animals, it is fair to consider whether the slight difference in tenderness is worth the cost. The answer is: perhaps not for many consumers.

When all animals are considered, there appears to be a relation between marbling and tenderness. Yet when the bias is removed by studying bulls and steers separately, there is no relation whatsoever between intramuscular fat and tenderness at 15 days. This, of course, points to the possibility that marbling is a sign of sex difference rather than tenderness of meat. It has been shown that some fat does contribute to the quality of juiciness (*5*), but the minimum fat requirements for juiciness have not been evaluated.

Carcass grade closely correlates with tenderness for the whole group. Yet when the steer bias is removed and steers and bulls are compared separately, the relationship between grade and tenderness disappears. This indicates that perhaps within a group of animals of similar breeding, age, sex, and feeding management, grade has little relation to tenderness.

Many factors are considered in assigning grades to carcasses and it would be desirable to know which contribute most to tenderness. Four have been considered here—sex, marbling, color, and age. The age of the animal at slaughter seems to contribute most to tenderness. All animals in this study were relatively tender and all were young and well fed. Although the age factor was not a primary variable in this study, it appears to be of major importance in spotting consumer quality in a carcass when compared in the light of previous reports from this laboratory (*7, 9, 10, 15*). Sex of the animal appears to be less important than age in indicating tender beef, and marbling of itself is of no importance except insofar as it may indicate sex differences. Color, as total pigment, seems not to be related to tenderness, except as it might contribute to the estimation of age and sex. All meat studied here was relatively constant in pH, and meat color as seen by the consumer is profoundly affected by pH, although total pigment may not be.

The factor of age is borne out by the lack of correlation between tenderness and hydroxyproline. The amount of connective tissue was low for all animals and all animals were relatively young and well fed. Consequently, contribution of connective tissue to the toughness of the animals studied here was relatively unimportant. In other animals it might well have been the dominant factor affecting tenderness.

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