

Connective Tissue in Beefburgers

DAVID W. LORD AND KAREN J. SWAN

County Laboratory, County Hall, Preston, Lancs

Data on the connective tissue content of meat products and meat are presented to facilitate the determination of lean meat in beefburgers. A maximum level of connective tissue content associated with lean meat in that product is recommended.

The Sausage and Other Meat Product Regulations 1967 required that meat with cereal should contain 80 per cent. of "meat" and 48 per cent. of "lean meat". "Lean meat" is not specifically defined but the "lean meat content" (as defined) of a product is to be made up of "lean meat free of visible fat". This is tantamount to saying that "lean meat" is *lean meat* and when free of visible fat must form 48 per cent. of a meat with cereal.

Thus the 1967 Regulations indicate a "technical" definition of lean meat which is—meat trimmed free of visible fat and containing only the connective tissue which would be normally associated with such trimmed meat. However, more importantly by effectively saying that "lean meat" is *lean meat* the Regulations leave the Courts to decide exactly what is lean meat. In this latter respect lean meat is a concept understood by the consumer. To the consumer lean meat is a piece of red muscle tissue without much visible fat and without much visible gristle. The new Meat Product Regulations continue this definition of "lean meat content", although a beefburger will be required to contain not less than 52 per cent. of "lean meat" as against 48 per cent. in the 1967 Regulations.

The term "connective tissue" is often used in a broad sense to indicate tissues which connect different parts of the muscles and, generally connect and hold bodily parts together^{2,3}. Thus individual muscle fibres are surrounded by endomysium, bundles of muscle fibres by perimysium and the muscle as a whole by epimysium. The tendon connects to the ends of the muscle which is linked to the external and internal connective tissues. In a cut of meat, "gristle" is likely to be thick streaks of perimysium or epimysium whilst the term "sinew" (used in the legislation) refers to tendon.

In addition the term "connective tissue" is extended to include other tissue such as skin, ligament and that of the aorta.

The connective tissues of the striated muscle described above are composed of the protein collagen (as in the skin)^{2,3,4,5,6}. Other connective tissues such as ligament and that of the aorta are composed mainly of the protein elastin^{3,7,8}.

Beef muscle, trimmed so that it forms lean meat, has associated with it the collagenous connective tissues described above. Clearly, a precise definition of

the maximum quantity of collagenous connective tissue is needed to enable the Official Analyst to check the lean meat content of a beefburger, or indeed, of any other meat product.

A review of the literature makes apparent a variety of methods of calculation of collagen or connective tissue content of meats. Many of them rely on determination of the hydroxyproline content of the tissue in question, accepting the fact that animal protein collagen has a uniquely high and fairly consistent level of hydroxyproline. The measurement of this amino acid is an indicator of the amount of connective tissue in a meat product.

It is essential that, in calculating the meat content of a beefburger, the relationship between lean meat, connective tissue and hydroxyproline is clearly defined.

It is suggested that there should be a defined maximum connective tissue content in the lean meat of a beefburger where connective tissue is expressed as:

$$\text{Wet-fat-free-connective tissue} = \text{Hydroxyproline} \times 37$$

and connective tissue nitrogen is expressed as

$$\text{Connective tissue nitrogen} = \text{Hydroxyproline} \times 1.28 \quad (1.28 \times 6.25) = 8$$

The factor 37 is one which was given consideration by the Meat and Meat Products Committee of the British Standards Institution⁹ some years ago.

It is a value judgement based on the following reasoning^{4,5,8,10,11,12,13,14,15,16,17}:

- (a) The hydroxyproline content of collagen is generally taken to be in the range 12 to 14 per cent;
- (b) Dry-fat-free-connective tissue contains around 85 per cent. of collagen; *- 10-12% if dry*
- and, (c) Wet-fat-free-connective tissue contains around 22.5 per cent. of protein.

This reasoning gives rise to the mathematical proposition:

$$\text{Dry-fat-free-connective tissue contains } \frac{14 \times 85}{100} = 12 \text{ per cent. hydroxyproline}$$

∴ Wet-fat-free-connective tissue contains

$$\frac{22.5 \times 12}{100} = 2.7 \text{ per cent. of hydroxyproline}$$

$$\begin{aligned} \therefore \text{Wet-fat-free-connective tissue} &= \text{hydroxyproline} \times \frac{100}{2.7} \\ &= \text{hydroxyproline} \times 37 \text{ per cent.} \end{aligned}$$

The expression for connective tissue nitrogen as hydroxyproline $\times 1.28$ reflects the range of hydroxyproline contents of collagen (i.e., 12-14 per cent. is equivalent to hydroxyproline $\times 7.1$ to 8.3) thus connective tissue protein = hydroxyproline $\times 8$. The factor 6.25 is used for conversion of connective tissue protein to nitrogen although it should be noted that lower factors have been suggested in the literature.

Thus

$$\text{Connective tissue nitrogen} = \frac{\text{hydroxyproline} \times 8}{6.25} = \text{hydroxyproline} \times 1.28.$$

It is worth noting that in Germany the following factors are prescribed¹⁸:

$$\begin{aligned}\text{Collagen nitrogen} &= \text{hydroxyproline} \times \frac{7.1}{5.55} \\ &= \text{hydroxyproline} \times 1.28.\end{aligned}$$

Combination of the use of these factors and the traditional Stubbs/More calculation for meat content¹⁹ allows the lean meat content of a beefburger to be calculated as outlined below.

CALCULATION OF LEAN MEAT CONTENT

$$\text{Wet fat free connective tissue} = \text{hydroxyproline} \times 37 = W$$

$$\text{Connective tissue nitrogen} = \text{hydroxyproline} \times 1.28$$

Fat free, connective tissue free, meat nitrogen =

$$\begin{aligned}\text{Total nitrogen} - (\text{connective tissue nitrogen} + \\ \text{other nitrogen (e.g. soya, carbohydrate)}) \\ = N\end{aligned}$$

$$\begin{aligned}\text{Connective tissue free, lean meat} &= N \times \frac{100}{3.55} \times \frac{100}{90} \text{ (i.e. assumes 10 per cent.} \\ &= L \text{ fat in "lean meat")}\end{aligned}$$

$$\text{Lean meat} = L \times \frac{100}{(100 - c)} = L_c \text{ (where } c = \text{the connective tissue assumed to be the maximum allowance for the lean meat).}$$

Where W is less than c , then lean meat = $L + W$ or when W is greater than c then excess connective tissue = $W - (L_c - L)$.

Clearly the use of the factors (37 and 1.28) and the general method of calculation are not beyond criticism and challenge. However it is more important that this paper presents:

- (a) the method of calculation of connective tissue content,
- and (b) results of measurements on meat and meat products using the defined method.

This will provide the basis for the conclusions reached.

Methods of Analysis and Quality Assurance

During the course of the laboratory work on raw meats, minced beef and beefburgers (reported later) the normal analytical procedures of the laboratory were employed. A minimum quality control rate of 10 per cent. was applied. This level is in accordance with the recommended protocol of the Association of Public Analysts²⁰. Brief descriptions of some of the quality control procedures applied are given below.

SUB-SAMPLING

For each category of meat and meat product sub-sampling was effected in two stages. Firstly, mechanical mincing to a progressively smaller particle size then homogenisation/blending until the sample was deemed to be of suitable homogeneity.

HYDROXYPROLINE

The method used was that of BS4401 Part II²¹.

Quality control was effected (a) by replication at a rate of 20 per cent. and (b) by checking the slope on each prepared calibration graph.

Replicates conformed to

Within Laboratory Repeatability Precision = 0.05 per cent.

(in the most common hydroxyproline range of 0.2 to 0.6 per cent.)

Note Within Laboratory Repeatability Precision is defined in the Association of Public Analysts protocol as:

the maximum absolute difference (95 per cent. probability) between two analytical results obtained by the same method in one laboratory. The repeatability precision is applicable to analytical results produced by different analysts on different days using different reagents etc.

TOTAL NITROGEN

A Kjeldahl method was used employing semi-automated block digestion and distillation.

Quality Control (at a rate of 20 per cent.) was effected by:

(a) Analysis of DL-Alanine standards conforming to a nitrogen recovery of

Within Laboratory Recovery Mean Value 99.5 per cent.

Within Laboratory Recovery Standard Deviation 0.5 per cent.

Note Within Laboratory Recovery terms are defined by the Association of Public Analysts protocol as follows:

Within Laboratory Recovery Mean Value is the mean recovery value (per cent.) calculated from a series of recovery experiments using the same method in one laboratory where recovery results are produced by different analysts on different days using different reagents etc.

Within Laboratory Recovery Standard Deviation is the standard deviation of the series of recovery experiments.

(b) Replication of nitrogen determinations on the meat products.

Replicates conformed to

Within Laboratory Repeatability Precision = 0.070 per cent. of

(in the most common nitrogen range of 2-3 per cent. of nitrogen)

SOYA PROTEIN

Approximately one third of the burgers contained soya protein. For analysis a combination of stereological technique²² and enzyme linked immunosorbent assay²³ was used. Quality control was effected by analysis of prepared meat/soya isolate/soya flour mixtures conforming to a soya recovery as follows

	Stereological	ELISA
Within Laboratory Recovery Mean Value	97	96
Within Laboratory Recovery Standard Deviation	9	7

Meat and Meat Product Surveys

Two cuts of cheaper meats, which have been known to be used in beefburger manufacture, have been examined and their connective tissue contents are shown in Table I.

TABLE I
CONNECTIVE TISSUE CONTENT OF TWO MEAT CUTS

Type	Description	Number of samples	Range of wet-fat-free-connective tissue contents per cent.
Shin	Retail cuts without further trimming	8	10 to 25
Diaphragm	Retail cuts without further trimming	5	2 to 7

In evaluating a reasonable level of connective tissue in beefburgers it is important to consider consumer expectation. A product used by consumers to prepare their own beefburgers is raw minced beef. The quality of that product gives some indication therefore of the quality of meat expected by the consumer in a beefburger. A survey of the connective tissue content of 31 samples of minced beef sold in butchers' shops, supermarkets etc. in Lancashire produced the data shown in Table II.

Beefburgers and "Beefburgers with Onion" have also been surveyed. A range of products purchased from retail outlets has been analysed and the results of 24 product analyses are shown in Table III.

Discussion

The Food Standards Committee in its Report on Meat Products²⁴ states:

For most cuts of meat, generally trimming should reduce the connective tissue content to less than 10 per cent. of the lean meat content. However slightly higher levels might occasionally be found even after trimming in cuts such as shoulder, flank, shin and brisket, which are the main cuts used for manufacturing.

This statement is supported by data on the connective tissue of meat before trimming.

The data indicates that before trimming the connective tissue content of cuts of beef lies typically in the range 10 to 19 per cent. After trimming, the connective tissue content will be significantly reduced, hence the Food Standards Committee recommendation that lean meat should be defined to include in most cases not more than 10 per cent. of connective tissue.

In effect the Food Standards Committee expects that in some meat products, depending on the cuts of meat used, the connective tissue content of those products will be a little above 10 per cent. but that upper level is not indicated.

Unfortunately the Food Standards Committee does not describe the basis of their calculation of connective tissue.

TABLE II
CONNECTIVE TISSUE CONTENT OF MINCED BEEF

	Wet fat free connective tissue expressed on product <i>per cent.</i>	Wet-fat-free-connective tissue expressed on apparent lean meat content* <i>per cent.</i>
	12	15
	20	21
	11	13
	15	15
	8	8
	20	20
	8	8
	17	20
	19	23
	14	16
	15	15
	14	14
	15	15
	15	15
	17	17
	10	12
	19	19
	20	22
	19	19
	20	22
	19	19
	18	18
	19	20
	14	16
	16	16
	16	17
	14	19
	12	13
	14	14
	13	13
	20	22
	18	18
	12	13
Maximum	20	23
Minimum	8	8
Mean	15	16

* Apparent lean meat content = $\left[\frac{\text{Nitrogen} \times 100}{3.55} \times \frac{100}{90} \right]$
or $\left[\frac{\text{Nitrogen} \times 100}{3.55} \right] + \text{fat content}$ whichever is the lower.

A survey of the literature reveals a significant amount of data on the hydroxyproline content of meat and meat products.

The data have been reported in many different ways but generally allow the hydroxyproline level in the original cut of meat to be calculated or postulated using average figures for meat composition²⁵. The hydroxyproline estimates have been converted to wet-fat-free-connective tissue content (by the defined method) in the analysis of the literature data shown in Table IV.

TABLE III
CONNECTIVE TISSUE CONTENT OF BEEFBURGERS

	Statutory required lean meat content claimed on label per cent.	Lean meat including 20 per cent. or less connective tissue per cent.	Connective tissue expressed on product per cent.	Connective tissue expressed on lean meat per cent.	Connective tissue in excess of 20 per cent. of the lean meat content per cent.
Major producers' national brands					
Beefburger	48	75	11	17	0
Beefburger	60	79	13	16	0
Beefburger with Onion	—	56	14	24	3
Beefburger with Onion	—	66	10	15	0
Beefburger	60	83	14	20	0
Beefburger	48	66	10	18	0
Beefburger	54	76	13	20	0
Beefburger	48	75	12	19	0
National "take-away/restaurant" products					
Beefburger	48	60	8	13	0
Beefburger	60	82	12	14	0
Major retailers' "own" brands					
Beefburger	48	68	11	19	0
Beefburger	48	70	12	20	0
Beefburger with Onion	—	60	18	28	6
Beefburger with Onion	—	55	9	17	0
Beefburger with Onion	—	66	9	14	0
Beefburger with Onion	—	70	10	14	—
Beefburger with Onion	—	61	10	16	0
Beefburger	48	67	11	16	0
Beefburger	48	67	10	14	0
Products from local butchers					
Beefburger	48	73	10	14	0
Beefburger	48	72	8	13	0
Beefburger	48	85	12	14	0
Beefburger	48	76	14	18	0
Beefburger	48	75	11	15	0
Maximum				28	
Minimum				13	
Mean				17	

A logical conclusion which can be reached from this is that many well trimmed cuts of meat contain less than 10 per cent. of connective tissue and in this respect the conclusions of the Food Standards Committee appear to be sound.

It is also apparent that meat products prepared from less well trimmed cuts (or from certain cuts having higher natural connective tissue levels) may have

TABLE IV
ESTIMATES FROM LITERATURE OF WET FAT FREE
CONNECTIVE TISSUE CONTENTS OF MEAT CUTS AND MEAT PRODUCTS

Meat type	Trimmed (T) or untrimmed (U)*	Number of cuts sampled	Number of samples	Estimate of wet-fat- free-connective tissue per cent.	Reference
Pork	T	9	9	up to 9	26
Beef	T	2	28	up to 5	12
Beef	T	2	8	up to 7	14
Veal	T	2			
Pork	T	2			
Lamb	T	2			
Beef offals	T	5	5	up to 10	15
Beef	T	2	40	up to 6	
Beef	T	5	90	up to 9	27
Veal	T	1	2	up to 11	8
Lamb/mutton	T	2	4	up to 16	
Beef	T	2	8	up to 15	
Beef	T	4	29	up to 9	28
Beef	T	1	11	up to 9	29
Beef	T	3	92	up to 8	30
Beef	U	8	8	up to 29	31
Corned beef	—	—	146	mean 15	17
Ham	—	—	25	mean 9	
Pork	—	—	22	mean 9	
Comminuted ham	—	—	11	mean 14	

* Methods of trimming described by the various authors varied but in many cases the removal of epimysium was indicated.

connective tissue contents above 10 per cent. In such products the lean meat (i.e. the well trimmed meat) may be expected to contain between 10 and 20 per cent. of connective tissue.

In Table I the data for shin beef are broadly in line with the Food Standards Committee's typical levels but Table I shows lower connective tissue levels for diaphragm.

Table II indicates that, on average, the consumer purchasing minced beef to make beefburgers would produce a burger which had 16 per cent. of connective tissue associated with the lean meat. It is worth noting that in many instances consumers do not expect the quality of minced beef to be high. The product is often regarded as being produced from trimmings or "left overs". Against this background a level of 16 per cent. connective tissue might not be perceived as being associated with a high quality product in the mind of the consumer.

It is also worth noting that in a study of frozen ground beef in France, Pailler¹⁶ recommended a quality guideline equivalent to 11 per cent. wet fat free connective tissue content.

In calculating the meat content of the burgers in Table III connective tissue levels up to a maximum of 20 per cent. of the lean meat have been assumed to be part of the lean meat (see calculation of lean meat content).

This figure of 20 per cent. is likely to be at the high end of the range of connective tissue which would reasonably be associated with the lean meat.

Seventeen of the products were beefburgers which were required to have a statutory lean meat content of between 48 and 60 per cent. (dependent on the claim made on their labels). All met their minima and none contained connective tissue in excess of 20 per cent.

Seven of the products were beefburgers with onion, none of which claimed a meat content on their label but they contained between 55 and 70 per cent. of lean meat. In two cases, after allowing the lean meat in the product to have associated with it 20 per cent. of connective tissue, the products have been described as containing excess connective tissue at levels of 3 and 6 per cent. However these products contained 56 and 60 per cent. of lean meat respectively, which are levels above the minimum for beefburgers of 48 per cent. Thus the amounts of excess connective tissue had no bearing on their minimum lean meat content requirement.

Table III shows that 22 of the 24 burgers contained lean meat which had associated with it 20 per cent. or less of connective tissue. The average connective tissue content expressed on the lean meat was 17 per cent.

Conclusions

The evidence presented from the literature and from analysis of meat cuts, minced beef and beefburgers indicates that it is reasonable to expect that a beefburger will contain lean meat having a connective tissue content (hydroxy-proline $\times 37$) between 10 and 20 per cent. It is recommended that not more than 20 per cent. of connective tissue (expressed as a proportion of the lean meat) should be allowed by the Official Analyst to form part of the lean meat content of the beefburger.

References

1. The Meat Product and Spreadable Fish Products Regulations, 1984, S.I. 1984, No. 1566. HMSO London, 1984.
2. Lawrie, R. *Developments in Meat Science—2*. Applied Science Publishers, London, 1981.
3. Etherington, D. J., and Sims, T. J., *J. Sci. Food Agric.*, 1981, **32**, 539.
4. Baker, L. C., Lampitt, L. H., and Brown, N. P., *J. Sci. Food Agric.*, 1954, **5**, 226.
5. Eastoe, J. E., *Biochem. J.*, 1955, **61**, 589.
6. Bailey, A. J., and Sims, T. J., *J. Sci. Food Agric.*, 1977, **28**, 565.
7. Neuman, R. E., and Logan, M. A., *J. Biol. Chem.*, 1950, **1984**, 299.
8. El-Hehyawi, M. G. E., *J. Vet. Sci. of UAR*, 1967, **4**, 25.
9. Private Communication.
10. Coomaraswamy, M., *J. Assoc. Publ. Analyt.*, 1972, **10**, 33.
11. Lee, Y. B., Elliot, J. G., Richansrud, D. A., and Hagberg, E. C., *J. Food Sci.*, 1978, **43**, 1359.
12. Ritchey, S. J., and Coven, S. J., *J. Agric. Food Chem.*, 1962, **10**, 40.
13. Bowes, J. H., *J. Soc. Leather Trades Chemists*, 1959, **43**, 203.
14. Aronson, J. N., and Elvehjem, C. A., *Food Res.*, 1956, **21**, 109.
15. Abdallah, N. M., El-Wakell, F. A., and Awad, A. A., *Eur. Vet. Med. J.*, 1979, **25**, 365.
16. Pailler, F. M., *Annales des Falsifications et de l'Expertise Chimique*, 1979, **72**, 776, 325.
17. Board, P. W., Montgomery, W. A., and Rutledge, P. J., *J. Sci. Food Agric.*, 1978, **29**, 569.
18. Prandl, O., Haas, J., and Polke, E., *Die Fleischwirtschaft*, 1967, **45**, 581.
19. Stubbs, G., and More, A., *Analyst*, 1919, **44**, 125.
20. Lord, D. W., *Analyt. Proc.*, 1984, **21**, 394.