N2,5

/seven

Fourth Meeting of European Meat Research Workers, Cambridge, September, 1958.

FACTORS AFFECTING THE WATER CONTENT OF MEAT

By

E. H. CALLOW

INTRODUCTION

It is not generally realised how many factors can affect the water content of meat. At the physiological level it can be affected by the proportion of fat present in the fatty and muscular tissues, and by the proportion of glycogen present in the muscular tissues. Because connective tissue contains a higher proportion of water than muscle fibre tissue, the proportion of connective tissue in muscle affects its water content. Moreover, it is possible to decrease the water-holding capacity of muscular tissue by fasting an animal, and to decrease it still further by allowing the fasting animal to drink water. When once the animal is killed, its carcase loses water by evaporation. Even frozen meat loses water by sublimation; but it may gain water during thawing, by condensation.

In the following paper an attempt will be made to evaluate the effect of these factors.

A. THE WATER CONTENT OF FATTY TISSUE

Fatty tissue consists of connective tissue cells engorged with fat (triglycerides). As fattening proceeds the percentage of fat (F/FT) increases, and the percentage of water (W/FT) consequently falls. Using data for the fatty tissues from a series of 10 ewes at all stages of fatness, the relation between W/FT and F/FT was given by the following equation:-

$W/FT = 88.5 - 0.890 F/FT \pm 0.4 (r = -0.9998)$

The analyses, which provided the data for this equation, were carried out under conditions which minimised any loss of water. Moreover, the figures used were the weighted averages for the fatty tissues as a whole from some/analyses for each carcase. Experimental error, therefore, was probably negligible.

Two things can be inferred from this equation.

1. The correlation co-efficient is not significantly different from 1.0. Hence the system is adequately described in terms of water (W), fat (F) and the dry, fat-free residue (R) of connective tissue which remains. Moreover

R/FT = 11.5 - 0.110 F/FT

2. It is implicit that the water-holding capacity of the connective tissue diminishes as the percentage of fat increases. This can be shown by calculating the water content on a fat-free basis (W/FTff). At 70% of fat, the value of W/FTff is 87.3%, decreasing to 86.5, 84.0 and 79.0% as the percentage of fat increases to 80, 90 and 95% respectively.

In another series of carcases (from 29 lambs) the data gave a similar equation.

 $W/FT = 82.0 - 0.826 F/FT \pm 0.4 (r = -0.9975)$

On this basis too the water on a fat free basis would decrease -

Again, the correlation coefficient is so near to 1.0 that the system may be described in terms of the three variables water, fat and dry fat free connective tissue.

With both these sets of data the loss of water has been minimised. In the case of the following data for the fatty tissue from beef animals there had been an appreciable loss of water from carcases by evaporation (see section C). Under these conditions the data gave the following equation: -

 $W/FT = 77.2 - 0.773 F/FT \pm 0.9 (r = -0.9977)$

Again the form of this equation is similar to the two previous ones; the constants, however, are smaller due to loss of water from the fatty tissues.

The general picture which emerges is that of fatty tissue consisting of connective tissue cells engorged with variable amounts of fat (triglycerides), and with the connective tissue becoming somewhat drier as fattening proceeds. In addition, of course, there can be a variable loss of water by evaporation from the connective tissue itself.

From time to time there emerges a suggestion that water can replace fat in fatty tissues when the reserves of fat are being used up. This cannot be substantiated because during a period of fasting the actual weight of the fatty tissue falls, and both fat (F) and water (W) and dry connective tissue (R) decrease in weight. In fattening too all three components W, F and R increase in weight, although F increases at a faster rate than either W or R.

B. THE WATER CONTENT OF MUSCULAR TISSUE

Anatomically, unlike fatty tissue, muscular tissue has a complicated structure. It consists of muscle fibre tissues supported by a soft skeleton of connective tissue, and interpenetrated by fatty tissue (marbling fat). Moreover, it may also have a sheath of connective tissue. The following discussion is limited to muscular tissue less any sheath.

Because of the presence of marbling fat, the percentage of fat (F/MT) increases. In the case of data for Psoas muscles for cattle (see eq. 1) and for lambs (see eq. 2) where the loss of water had been minimised the relation was given by:-

1. $W/MT = 79.6 - 0.963 F/MT \pm 0.4 (r = -0.966)$

2. $W/MT = 79.3 - 0.989 F/MT \pm 0.7 (r = -0.885)$

/13

Data for the Rectus femoris muscle from 10 ewes at all levels of fatness gave the following relation: -

 $W/MT = 79.4 - 0.854 F/MT \pm 0.8 (r = -0.905)$

When the data for the weighted averages for F/MT and W/MT (based on/analyses for each carcase) for the total muscular tissue from a series of 28 lambs were used the equation was: -

$$W/MT = 79.5 - 0.926 F/MT \pm 0.5 (r = -0.965)$$

And when similar data for the total muscular tissue from a series of beef carcases were used the equation was: -

80

- 3 -

Here the decrease in the value for the constants isdue to a loss of water by evaporation from the carcases (see section C).

One conclusion is permissible from these equations, namely that the description of muscular tissue in terms of fat (F), water (W) and dry fat-free tissue (R) is not adequate in a mathematical sense, as it was with fatty tissue. The reason for this becomes apparent when we consider the changes in the anatomy of a single muscle in relation to growth and fattening. Data illustrating these changes for the Rectus femoris muscle from a series of 10 adult ewes at all levels of fatness are given in Table I.

No.	Weight of muscle	Average diameter of muscle fibres	Percentage of connec- tive tissue	Percent -age of fat	Percent -age of Water	Percentage of water on a fat- free basis
	MT	D M	CT/MT	F/MT	W/MT	W/MT _{ff}
11 21 26 20 10 9 29 4 30 12	181 249 276 309 316 339 372 411 426 484	33.9 38.7 42.6 42.5 43.0 46.4 47.0 46.4 47.7 52.4 45.3	15.2 9.7 13.4 10.3 11.6 11.2 7.8 8.1 7.4 8.8	0.7 1.8 2.2 3.8 4.4 7.9 5.1 3.7	79.8 77.9 77.7 75.5 76.5 74.4 74.5 74.7 75.9 75.7	80.3 79.3 79.5 78.5 80.1 78.4 78.7 78.7 78.3 78.7

TABLE I

The general picture that emerges from these data is that an increase in the weight of the muscle (MT) is accompanied by an increase in the diameter D of the muscle fibres as shown by the following equation: -

 $D = 28.1 + 0.0475 \text{ MT} \pm 3.0 (r = +0.838)$

Moreover, the increase in weight is associated with a decrease in the percentage of connective tissue and an increase in the percentage offat. Because of the decrease in the percentage of connective tissue there is a decrease in the percentage of water in the fat-free tissue.

 $W/MT_{ff} = 76.6 + 0.25 CT/MT \pm 0.4 (r = +0.83)$

For this reason, and possibly because, as we have seen, connective tissue itself appears to hold less water as fattening proceeds, the water content of muscular tissue, when calculated on a fatfree basis decreases during fattening. Thus if calculations are made with the aid of equation I. it can be calculated that W/MTff is 78.9% when F/MT is 4% and only 78.1% when F/MT is 8%.

It follows from this evidence that the water content of muscular tissue (calculated on a fat-free basis) should be less in muscles from fat animals than it is in those from lean animals. Evidence for this has been obtained by studying the water content of Psoas muscles from 29 beef animals whose carcases contained from 7.5% to 39.4% of fatty tissue (FT/C). The percentage of water on a fat-free basis ($W_{\rm ff}$) from these muscles was negatively correlated, at a very high level of significance, with the fatness of the carcases as is shown by the following equation:-

$W_{ff} = 80.4 - 0.0929 FT/C + 0.77 (r = -0.755)$

Moreover, within a single carcase we may expect to find some muscles (those containing more connective tissue) with a high water content (on a fat-free basis) than others (those containing less connective tissue). Evidence for this was obtained from analytical data for the rectus femoris and the psoas muscles from 29 lambs. The average value was 78.8% for the rectus femoris muscle (with high content of connective tissue), and 78.4% for the psoas muscle (with a low content of connective tissue). This difference was statistically significant at the highest level. Moreover, the variation from animal to animal (as is implicit in the previous equation) was also statistically significant.

It is, thus, clear that the overall water content of muscular tissue is increased by the presence of connective tissue, and decreased by the presence of fat. In a similar way we may expect the presence of glycogen to decrease the overall percentage of water in muscular tissue. In this connection it is important to realise that the glycogen present in living muscular tissue can vary considerably and amounts between 0.8 and 2.5% have been recorded. (Howard and Lawrie, 1956).

The effect of fasting. There is some evidence that the waterholding capacity of muscular tissues can be affected by the level of nutrition of the live animal. Thus, with pigs, there is a surprisingly rapid loss of weight of the tissues of the carcase if they are deprived of food. Work carried out in 1936 showed that this loss (in a 200 lb. live weight pig) was about 3 lbs. of carcase weight per 24 hours. Moreover, if the pigs had been given water, there was an extra loss of 4 lbs. per carcase. Calculations showed that even the 3 lbs. of carcase wastage could not be attributed entirely to normal wastage of muscular tissues and of fatty tissues (Callow and Duckham, 1937p54). Whereas a bacon pig might be expected to produce 5,000 cals. by metabolic processes in one day, a loss of 3 lbs. would indicate 9,000 cals. if it were due to tissue wastage alone. The suggestion was, therefore, made that some of the wastage was due to a loss of water from the remaining tissues. Evidence for this was obtained by analysing the Pscas muscles of two groups of 48 pigs, one group of which had been partially fasted for two days. The control group had Pscas muscles which contained on the average 76.3% of water and the fasted group had muscles which contained 75.9% of water. This result, although significant (p = .01), is not of sufficient magnitude to account for the losses recorded above. It should he noted, however, that fasting was only partial.

The mechanism of the loss of capacity to hold water by muscular tissue has still to be established. It is, however, known that tissue cells contain more K ions than are found in the lymph which surrounds them. Moreover, energy has to be provided to maintain this "steady state". If the water holding capacity of muscular fibres is dependent on this steady state, it becomes clear how fasting and even the ingestion of water could reduce it.

Although it is hard to obtain evidence for the relation between energy intake and water holding capacity in the case of muscular tissue, there is no doubt about it in the case of pigs' livers. In an experiment where two pigs were fasted for 24 hours

88

$$W_{PP} = 80.4 - 0.0929 FT/C \pm 0.77 (r = -0.755)$$

Moreover, within a single carcase we may expect to find some muscles (those containing more connective tissue) with a high water content (on a fat-free basis) than others (those containing less connective tissue). Evidence for this was obtained from analytical data for the rectus femoris and the psoas muscles from 29 lambs. The average value was 78.8% for the rectus femoris muscle (with high content of connective tissue), and 78.4% for the psoas muscle (with a low content of connective tissue). This difference was statistically significant at the highest level. Moreover, the variation from animal to animal (as is implicit in the previous equation) was also statistically significant.

It is, thus, clear that the overall water content of muscular tissue is increased by the presence of connective tissue, and decreased by the presence of fat. In a similar way we may expect the presence of glycogen to decrease the overall percentage of water in muscular tissue. In this connection it is important to realise that the glycogen present in living muscular tissue can vary considerably and amounts between 0.8 and 2.5% have been recorded. (Howard and Lawrie, 1956).

The effect of fasting. There is some evidence that the waterholding capacity of Muscular tissues can be affected by the level of nutrition of the live animal. Thus, with pigs, there is a surprisingly rapid loss of weight of the tissues of the carcase if they are deprived of food. Work carried out in 1936 showed that this loss (in a 200 lb. live weight pig) was about 3 lbs. of carcase weight per 24 hours. Moreover, if the pigs had been given water, there was an extra loss of 4 lbs. per carcase. Calculations showed that even the 3 lbs. of carcase wastage could not be attributed entirely to normal wastage of muscular tissues and of fatty tissues (Callow and Ducham, 1937p54). Whereas a bacon pig might be expected to produce 5,000 cals. by metabolic processes in one day, a loss of 3 lbs. would indicate 9,000 cals. if it were due to tissue wastage alone. The suggestion was, therefore, made that some of the wastage was due to a loss of water from the remaining tissues. Evidence for this was obtained by analysing the Psoas muscles of two groups of 48 pigs, one group of which had been partially fasted for two days. The control group had Psoas muscles which contained on the average 76.3% of water. This result, although significant (p = .01), is not of sufficient magnitude to account for the losses recorded above. It should he noted, however, that fasting was only partial.

The mechanism of the loss of capacity to hold water by muscular tissue has still to be established. It is, however, known that tissue cells contain more K ions than are found in the lymph which surrounds them. Moreover, energy has to be provided to maintain this "steady state". If the water holding capacity of muscular fibres is dependent on this steady state, it becomes clear how fasting and even the ingestion of water could reduce it.

Although it is hard to obtain evidence for the relation between energy intake and water holding capacity in the case of muscular tissue, there is no doubt about it in the case of pigs' livers. In an experiment where two pigs were fasted for 24 hours

88

before slaughter, and where two pigs were given a meal containing 2 lbs. of sugar three hours before slaughter and another pair two such meals, one 18 hours and the other 3 hours before slaughter, the livers showed a progressive increase in weight (see Table II).

TABLE II

The weight of livers, and of water, carbohydrate, protein and fat in these livers of six pigs subjected to three levels of nutrition immediately before slaughter.

Treatment of live animal	Weight of liver gms.	Weight of water gms.	Weight of carbo- hydrate gms.	Weight of protein, etc. gms.	Weight of fat gms.
24 hours fast	1084 1212	747 830	4 6	241 273	92 103
24 hours fast plus one meal	1301 1383	939 984	42 63	256 283	64 53
24 hours fast plus two meals	1721 2231	1230 1583	162 279	316 349	13 20

Moreover, there was a progressive increase in weight of water, carbohydrate and protein. Oddly enough, there was a progressive decrease in the content of fat. These changes were very closely related to the actual weight of the livers, as is shown by the constants etc. for the regression lines relating the weight of the livers with the weight of water etc. in the livers. (See Table III).

TABLE III

The constants for the regression equations relating the weight of water, carbohydrate, protein and fat in livers with the total weight of liver (L). The generalised form of equation used being, Weight of part = $a + bL \pm C$.

Weight of	a	Ъ	<u>+</u>	r
Water	- 31	+0.7275	18.3	+0.9986
Carbohydrate	-286	+0.2545	12.2	+0.9949
Water + Carbohydrate	-317	+0.9820	7.7	+0.9986
Protein etc.	+150	+0.0914	11.4	+0.9666
Fat	+167	-0.0734	21.8	-0.8464

p is less than .001 for all equations except that for fat where it lies between .01 and .02.

Two things are quite clear from these data: 1) the increase in weight of the livers is almost entirely due to the increase in weight of carbohydrate and water - over 98% of the increase is accounted for in this way. 2) The ratio of water to protein increases steadily with the increase in weight of the liver. From these data the inevitable conclusion is that livers increase in weight mainly by uptake of carbohydrate and associated water. Moreover, a given weight of carbohydrate is not associated with a given weight of water. The effect is, therefore, a physiological one and not a chemical one.

With regard to muscular tissue, however, all that is claimed is that its water content is decreased by a short period of fasting and that the ratio of water to protein is decreased.

C. LOSS OF WATER FROM CARCASE MEAT

So far the emphasis has been on the physiological factors which influence the water content of meat. We now come to the effect of physical factors.

When once a carcase is exposed to the air it begins to lose weight by the evaporation of water. For this reason "cold dead weight" is always appreciably less than "hot dead weight" and in bacon factories a deduction of 4 lbs. from a carcase weighing 150 lbs. "hot" is allowed for this loss during cooling.

It might be expected that this loss would depend on the fatness of the meat, and that leaner meat would lose more water than would fat meat. In the case of sides of beef left to cool overnight at room temperature I this relation was found to hold, and the percentage loss in weight of the side (L/S) was found to be significantly correlated (p = .02) with the percentage of fatty tissue (FT/S) in the side, as may be seen from the following equation: -

 $L/S = 2.67 - 0.0387 FT/C \pm 0.38 (r = -0.64)$

It would, however, be truer to relate this loss in weight with the percentage of water in the side (W/S). When this is done the following relation, which is a more significant one (p = .01), holds: -

L/S = 0.0984 W/S - 2.92 + 0.35 (r = +0.709)

From this it can be calculated that the loss of water from a lean side (containing 50% of water) would be 2%, whereas from a reasonably fat side (containing 40% of water) it would only be 1%.

In practice this loss of weight by evaporation can be reduced by hanging the carcase meat in a chill room straight away and by controlling the conditions of temperature, humidity and air movement in the chill room. Thus with bacon sides which were cooled at room temperature overnight and then chilled for 24 hours, a loss of 0.7 lbs. (on a 60 lb. side) was reduced to 0.4 lbs. by immediate chilling. Even this loss can be reduced if the air is kept more humid and the air speed diminished.

Even frozen meat loses weight by the sublimation of ice to water vapour. Thus during the transport by ship of frozen lamb carcases from New Zealand to England losses of the order of 0.5% were recorded/by a further 1.2% during cold storage on land for a month. Such losses of weight occur almost entirely at the surface of the meat and, should they be excessive, a porous structure is left by the sublimed ice; a condition known as "freezer burn".

I The animals were killed over a period from March to September. Climatic conditions - and hence rates of evaporation - must thus have varied considerably.

- 6 -

Finally, when frozen meat is thawed, water is condensed on its surface and, during the first hour of exposure of frozen lamb carcases to air at 17°C, they have been known to gain 0.6% in weight as a consequence.

D. THE WATER CONTENT OF COMMERCIAL MEAT

From what has been said, it might appear that any attempt to define the water content of meat was doomed to failure. There is, however, no need to take such a gloomy point of view. If we define meat as the fatty and muscular tissues removed from a carcase at a stage when it has lost about 2 to $2\frac{1}{2}\%$ of its weight by evaporation, i.e. meat as the butcher handles it, then the following data have been established.

In a study of 22 beef carcases (all containing more than 18% of fatty tissue) it was found that, on a fat-free basis, the water content of the muscular tissue was $77.1 \pm 0.3\%$ and that of the fatty tissues $76.9 \quad 1.5\%$ (see Table IV). Moreover, the relation between the percentage of fat (F/BM) and the percentage of water (W/BM) in the boneless meat, i.e. the combined muscular and fatty tissues, was given by the equation: -

$W/BM = 77.0 - 0.766 F/BM \pm 0.2 (r = -0.9994)$

The constant 77.0 in the above equation in fact represents the value when F/BM is zero - i.e. it represents a value for water on a fat-free basis.

If the American food tables prepared by Chatfield Adams be consulted the water content of beef, veal, lamb and pork on a fat-free basis can be calculated, and is found to be 77.0 ± 0.5 . If the value of 77.0% of water on a fat-free basis be accepted as a standard figure for boneless meat, then the relations between fat, water and protein are also standardised.

TABLE IV

	Reserved of the second s	
The (on	e chemical composition of boneless meat and the w n at fat-free basis) of the muscular and fatty ti	ater-content .ssues of beef animals

$\begin{array}{c c c c c c c c c c c c c c c c c c c $						and the second sec			and the support of the same in the	Particular and the second s
79.6 77.0 83.4 73.5 76.7 77.7 77.1 80.0 74.8 77.1 77.2 77.0 81.1 77.8 77.1 78.9 77.0 85.0 77.5 77.0 77.9 76.9 81.6 75.3 77.5 77.3 76.9 81.1 75.8 76.8 77.6 77.7 83.6 75.3 77.5 77.6 77.0 85.5 79.7 77.2 77.6 77.0 85.8 77.4 77.0 77.5 77.0 82.2 76.9 77.4 77.5 77.2 84.8 79.0 77.4 77.5 77.2 84.8 79.0 77.4 77.5 77.2 84.8 79.0 77.4 77.5 77.2 80.2 76.9 77.4 77.5 77.2 79.6 76.5 76.4 76.7 77.1 80.7 79.1 77.5 77.6 77.7 73.5 74.9 77.5 77.6 77.7 80.8 78.3 76.9 77.1 76.9 77.2 80.8 78.5 77.4 77.6 77.2 79.8 76.7 77.6 77.5 76.8 86.4 77.1 77.9 77.6 77.2 80.8 79.0 77.4 77.6 77.2 79.9 75.6 77.4 77.6 77.2 84.8 79.0 77.4 <t< th=""><th>Pamu</th><th>soas iscles</th><th>Mu - ti</th><th>iscula ssues</th><th>r Peri fatt</th><th>nephric y tissue^I</th><th>Fati</th><th>tty ssues</th><th>Bo</th><th>ne-less meat</th></t<>	Pamu	soas iscles	Mu - ti	iscula ssues	r Peri fatt	nephric y tissue ^I	Fati	tty ssues	Bo	ne-less meat
10.3 14.2 1.3 10.3	age	79.6 777.2 99.3 77.2 77.7 77.7 77.7 77.7 77.7 77.7 77		77.0 77.1 77.0 77.0 77.0 77.0 77.0 77.0		83.4 80.0 81.1 85.0 81.6 83.6 83.1 85.5 84.8 85.8 84.2 79.0 79.4 80.2 79.0 76.4 79.0 80.2 780.8 80.2 780.8 80.2 780.8 80.2 780.8 80.2 79.8 80.2 79.8 80.2 80.0 80.2 79.8 80.2 80.0 80.2 79.0 80.2 79.0 80.2 79.0 80.2 79.0 80.2 79.0 80.2 80.2 80.0 80.2 80.0 80.0 80.0 80		73.5885338740969511937156095		76.7 77.9 77.08 77.6.85 77.6.85 77.6.82 77.6.82 77.6.85 77.77 77.77 77.6.85 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.6.9 77.77 77.99 9.40 77.10

I These values were determined as soon after slaughter as possible and losses due to evaporation were minimised. The percentage of fat (F/BM), water (W/BM) and the remainder, i.e. the dry fat residue (R/BM) must add up to 100. But the relation between the water and the dry fat-free residue must be as 77:23 if water on a fat-free basis is to be 77%.

In the equation:

F/BM + R/BM + W/BM = 100

R/BM can now be put as $\frac{23W}{77}$ /BM

92

The equation can now be re-written as:

W/BM = 77-0.77 F/BM

In Fig. 1 the straight line has been drawn to represent this equation. It will be seen that our own deta for boneless beef and the American data for beef, veal, lamb and pork fit it remarkably well, except for boneless meat containing less than 10% of fat. With such lean animals, as we have already seen, the muscular tissue contains more connective tissue and hence more water than does that from fatter animals.

One further point, the equation for boneless beef is almost identical with the one represented by the straight line in Fig. 1. The constant is 77.0, and although the slope is .766 this is not significantly different from .770. (In fact a calculation of the "t" test for the difference gives t = .7 which gives a value p = .5, i.e. the odds are even that the slope of the two lines is identical.) We may, therefore, accept the following equations as good approximations.

> W/BM = 77.0 - 0.77 F/MR/BM = 23.0 - 0.23 F/M

If the further assumption is made that 90% of the dry fat-free residue (R) is protein (P), then:

P/BM = 20.7 - 0.207 FM.