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A PROTOCOL TO MEASURE THE FREE WATER IN RAW AND COOKED MEAT

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Abstract – The water holding capacity of meat is one of the most important factors affecting the quality. There is a multitude of procedures to measure the WHC on raw and cooked meat but results in the literature are difficult to compare and correlate. To solve this problem it has been developed a protocol to be used on the same meat sample, to analyze the water loss at different stages of its commercial food life during: thawing, cooking, cooling and consumption. The water on raw meat is measured as: thawing; drip; total area, ring and meat film area at 10' by compression; free water. On cooked meat: cooking and cooling loss; available cooked meat water. Parameters are expressed as the per cent out of the total water content. The traditional parameters are achieved at a lower cost and with coefficients of variation lower or at most similar to that reported in the literature. In addition, two new parameters which measure cooling loss and available cooked meat water allow to better define the available water at the time of consumption. The protocol uses two devices specifically designed by the authors to automate and improve the accuracy of some of the proposed parameters.

I. INTRODUCTION

Meat has a central role in our eating and the world consumer's demand is still increasing. The water holding capacity (WHC) of meat and meat products is one of the most important factors affecting economic value and meat quality. WHC affects the weight change during transport, storage, thawing, weight loss and shrinkage during cooking, and juiciness and tenderness of the meat (1). The WHC is the ability of meat to retain its water during application of external forces, such as cutting, grinding or pressing and processing (1). Changes in WHC in muscle *post mortem* involves lateral shrinkage of myofibrils as the principle mechanism driving water loss but there are many other factors, both genetic and non-genetic, that can influence WHC. There is a multitude of procedures to measure the WHC on raw and cooked meat or meat products (2, 3, 4, 5, 6). Because of the variation in used methods, the results for the water loss in the literature are difficult to compare. All the methods chosen to measure water loss in raw and cooked meat are generally independent of each other. Each method requires its sample and the treatment is specific but this includes an assumption of homogeneity of the muscle under investigation, an assumption that is particularly critical when measuring drip loss (7). So in addition to the method variability there is the variability of the meat samples. The increased variability reduces the correlation among the different methods that measure the loss of water at different times of commercial life and during consumption. To solve this problem it has been developed a protocol to be used on the same fresh or frozen meat sample, to analyze the water loss at different stages of his simulated commercial food life during: thawing, ageing, cooking, cooling and consumption.

II. MATERIALS AND METHODS

The proposed protocol measures water loss on raw meat as: thawing (TH); drip (DL); Water Holding Capacity (whc600), ring (ring), the ratio meat film area on total area (whcrp) at 10'; the free water (frwt). On cooked meat: cooking (clmcs) and cooling (cwmcs) loss by Meat Cooking Shrinkage (MCS) method (8); water loss (srwl) by Stress Resistance and Relaxation (SRR) method (9). It is also reported the total moisture content of the meat (tmc) and the pH at the analysis. The parameters TH, DL, frwt, clmcs cwmcs and srwl are expressed as the per cent out of the total water content to compare the contribution of each parameter. All these analysis are carried out, in a consecutive way, using only a 3cm thick sample (*M. Longissimus thoracis*). The showed results were obtained from the analysis of samples get out from meat aged for 7 to 14 days and fresh or frozen.

Meat sample

M. Longissimus thoracis obtained from veal, beef and pork were used as source of samples from the 12th thoracic rib in the caudal direction. When frozen the samples were vacuum packed and stored at -20°C until analysis. Two days before analysis day sample were thawed at 4°C.

Thawing loss (TH)

The day of analysis every thawed sample was taken from the bag and weighed. The bag was gently dried using blotting paper and then it was weighed. The difference between frozen packaged sample *minus* the thawed sample and the bag was the thawing loss. It was expressed as the per cent out of the weight of the frozen packaged sample and corrected for the tmc.

Drip Loss (DL)

A first slice (1cm thickness) was taken from the sample. A rectangular meat piece (4x4cm) was obtained from this slice. It was weighed and put in a plastic closed container. This container had a perforated support that permitted the escape of fluid. After a storage period (48h) at chill temperature (4°C) sample was weighed again (4). The difference between initial weight and the weight after storage was DL measurement. It was expressed as the per cent out of the initial weight and corrected for the tmc.

Water Holding Capacity (whc600), meat film area/whc600 (whcrp) and Ring

Raw meat remains (80g), obtained from the previous analysis, were trimmed of external fat and chopped before grinding. Afterwards 250 ± 10mg of minced meat were weighed on a filter paper sheet to measure total and meat film area (whcmt) after 600s of compression at 500N (10). Three replicates of every sample were carried out. The total area in mm^2 on the filter paper was the Water Holding Capacity at 10' (whc600) and the difference to the whcmt, expressed as the per cent out of the whc600, evaluates the ring. The whcrp measures the whcmt as the per cent out of the whc600.

Free water (frwt)

The free water is the ring area expressed as the per cent out of the total moisture content (tmc) according to Wierbicki and Deathrage (11) as reported in the formula [1] where 0.09470518941 mg of water/mm^2 is the regression coefficient.

[1] $$frwt = \frac{(whc600 - whcmt) * 0.09470518941}{weight\ sample * H_2O}$$

Total cooked loss by MCS (clmcs)

A second slice (1 cm of thickness) was cut from initial sample using a knife and a cutting system, based on two horizontal guides. From this slice a circular steak (5.5cm Ø) was obtained according to MCS protocol (8). Row sample, on temperate glass, was weighed then cooked in an electric forced air convection oven for 600s at 165°C and 69-70°C in the center of the sample. After it was put in a Petri dish, between two dried filter paper sheets. After 20min of cooling at room temperature, the circular steak was gently dry and weighed. The difference among raw and cooked sample was the cooked loss (clmcs), expressed as the per cent out of the raw sample weight and corrected for the tmc.

Cooling loss by MCS (cwmcs)

Petri dish with two dry filter paper sheets was weighed before and after sample cooling. The difference expressed as the per cent out of the raw sample weight, evaluates the cooling water loss (cwmcs) and corrected for the tmc. Data didn't need to be corrected for the expressible fat as it was irrelevant according to our test.

Cooking loss by MCS (clwmcs)

The difference between the total cooked loss (clmcs) and the cooling loss (cwmcs) measures the water lost in the oven when meat is cooking, expressed as the per cent out of the clmcs (8) and corrected for the tmc.

Available cooked meat water by SRR (srwl)

The cooled previous circular cooked steak was used to obtain three cylinders of 1cm of diameter. These cylinders were compressed according to the SRR method (9). Each cylinder was weighed before and after compression and the difference, expressed as the per cent of the cooked cylinder, evaluates the water in the cooked meat available to the consumer (srwl) and corrected for the tmc.

Statistical analysis was carried out with the SAS version 9.4 (12) using the Pearson Correlation analysis among continuous variables. Results are expressed as Means, standard deviations (STD) and coefficients of variation.

III. RESULTS AND DISCUSSION

In Table 1 are reported the means and their coefficients of variation. The N is variable according to the availability of the measured parameters. The coefficients of variation are very large for some parameters also due to effect of species and categories. Muchenje *et al.* (13) reported similar ranges for some parameters related to the WHC: drip loss 0.14-3.89%; WHC 37.0-72.7%; cooking loss 13.1-34.5%. Otto *et al.* (5) reported, for the drip loss using two different methods, a coefficient of variation ranging between 47.4 to 65.1%. Cheng *et al.* (14) reported coefficients of variation for drip loss of

Table 1 Mean, STD and Coefficient of variation of the water loss parameters (%).

Parameters	N	Mean	STD	CV
Thawing loss	226	7.84	3.68	46.9
Drip loss	285	5.30	3.57	67.4
WHC (mm^2)	407	1363.8	86.2	6.3
Ring	407	41.66	6.92	16.6
Meat film area/Total area	407	58.34	6.92	11.9
Free water	325	28.23	5.73	20.3
Total cooked loss	332	30.50	5.94	19.5
Cooling loss	308	5.81	2.18	37.5
Cooking loss	308	24.72	6.40	25.9
Available cooked meat water	116	35.30	7.34	20.8
Total moisture content	334	74.13	2.10	2.8
pH	121	5.47	0.22	4.0

63.5%, cooking loss 31.5% and cooling loss 43.4%.

The measured parameters by the proposed protocol analyze all the different water content in the raw and cooked meat. It is possible to analyze how the meat water is lost during the shelf life until consumption.

The maximum water loss is during cooking meat (24.72% out of tmc) and when cooling the meat still loses 5.81% out of tmc for a total cooked loss of 30.50%. The available cooked meat water (srwl) is more than a third (35.3 out of tmc). The sum of the parameters measured on the cooked meat is equal to 65.83% plus the thawing loss (7.84%) and the drip loss (5.3%) is equal to 79% which is not the expected free water (85-95%) (15). The missing 6-16% of free water could be in the residual sample at the end of the SRR method. This method compresses the 1cm cylinder to 0.75cm, and then there is still water in the sample.

The free water measured by compression on the raw meat was 28.23% out of the tmc and summed to TH and DL is equal to 41.4%, very far from the expected free water (85-95%). It was added also the DL but it should already be considered in the measured free water.

The table 2 shows the correlation among parameters. The TH is negatively correlated to the DL, whc600 and cwmcs; positively to the clmcs and clwmcs. Less water is lost during thawing and greater is the amount lost later on raw meat but not on the cooked meat. DL is positively correlate with frwt, whc600 and ring to indicate that higher is the free water and greater will be the losses. DL also indicates a greater loss on cooked meat. DL is negatively correlated to the whcrp which indicates a larger percentage of the meat film area, then less available free water. DL is negatively correlated to the pH to confirm what is already known (15, 16). The whc600, ring and frwt are positively correlated to the other parameters except TH, cwmcs and pH. The whcrp is negatively correlated to them because it measures the meat film area.

The clmcs is negatively correlated to the srwl to indicate a lower loss when cooking and cooling then more water when the meat is consumed. The tmc is positively correlated to the compression methods on the raw meat (whc600, ring and frwt) and, as expected, negatively with whcrp. It is not clear why it was found a strong negative correlation between pH and srwl (-0.55); it would be expected a positive correlation.

IV. CONCLUSION

The proposed protocol achieves some of the traditional parameters at a lower cost and with coefficients of variation lower or at most similar to that reported in the literature. In addition, two new parameters which measure cooling loss and available cooked meat water allow to better define the water available at the time of consumption. It is responsible, along with the fat content, of the sensation of juiciness detected by the consumer. The protocol uses two devices specifically designed by the authors to automate and improve the accuracy of some of the proposed parameters.

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Table 2. Correlations among water loss parameters

Pearson Correlation Coefficients
Prob > |r| under H0: Rho=0
Number of Observations

	TH	**DL**	**whc600**	**ring**	**frwt**	**whcrp**	**clmcs**	**cwmcs**	**clwmcs**	**srwl**	**tmc**	**pH**
TH Thawing Loss	1.00000 226	-0.40308 <.0001 183	-0.12097 0.0747 218	-0.11268 0.0970 218	-0.10973 0.1062 218	0.11268 0.0970 218	0.15651 0.0188 225	-0.22320 0.0010 213	0.23015 0.0007 213	-0.15650 0.2728 51	-0.10772 0.1063 226	0.16897 0.4093 26
DL Drip Loss	-0.40308 <.0001 183	1.00000 285	0.21920 0.0002 279	0.21550 0.0003 279	0.22819 0.0001 279	-0.21550 0.0003 279	0.31365 <.0001 284	0.19007 0.0020 262	0.23011 0.0002 262	-0.10658 0.2569 115	0.01847 0.7562 285	-0.38215 0.0448 28
whc600 10' WHC Total Area	-0.12097 0.0747 218	0.21920 0.0002 279	1.00000 407	0.61809 <.0001 407	0.79981 <.0001 325	-0.61809 <.0001 407	0.30055 <.0001 324	0.05618 0.3321 300	0.25759 <.0001 300	0.18221 0.0556 111	0.35592 <.0001 325	-0.12174 0.2790 81
ring Ring water	-0.11268 0.0970 218	0.21550 0.0003 279	0.61809 <.0001 407	1.00000 407	0.96001 <.0001 325	-1.00000 <.0001 407	0.29920 <.0001 324	0.01692 0.7703 300	0.26391 <.0001 300	0.08809 0.3579 111	0.31399 <.0001 325	0.00948 0.9330 81
frwt Free water %	-0.10973 0.1062 218	0.22819 0.0001 279	0.79981 <.0001 325	0.96001 <.0001 325	1.00000 325	-0.96001 <.0001 325	0.33822 <.0001 324	0.03300 0.5691 300	0.29834 <.0001 300	0.11239 0.2402 111	0.23198 <.0001 325	-0.11543 0.5912 24
whcrp Meat area/total %	0.11268 0.0970 218	-0.21550 0.0003 279	-0.61809 <.0001 407	-1.00000 <.0001 407	-0.96001 <.0001 325	1.00000 407	-0.29920 <.0001 324	-0.01692 0.7703 300	-0.26391 <.0001 300	-0.08809 0.3579 111	-0.31399 <.0001 325	-0.00948 0.9330 81
clmcs Total Cooked Loss by MCS	0.15651 0.0188 225	0.31365 <.0001 284	0.30055 <.0001 324	0.29920 <.0001 324	0.33822 <.0001 324	-0.29920 <.0001 324	1.00000 332	-0.03624 0.5263 308	0.94050 <.0001 308	-0.26650 0.0038 116	-0.08352 0.1288 332	-0.20919 0.2761 29
cwmcs Cooling Loss by MCS	-0.22320 0.0010 213	0.19007 0.0020 262	0.05618 0.3321 300	0.01692 0.7703 300	0.03300 0.5691 300	-0.01692 0.7703 300	-0.03624 0.5263 308	1.00000 308	-0.37366 <.0001 308	-0.12239 0.1906 116	-0.10458 0.0668 308	-0.08621 0.6566 29
clwmcs Cooking loss by MCS	0.23015 0.0007 213	0.23011 0.0002 262	0.25759 <.0001 300	0.26391 <.0001 300	0.29834 <.0001 300	-0.26391 <.0001 300	0.94050 <.0001 308	-0.37366 <.0001 308	1.00000 308	-0.20041 0.0310 116	-0.07120 0.2127 308	-0.19614 0.3079 29
srwl Available cooked meat water	-0.15650 0.2728 51	-0.10658 0.2569 115	0.18221 0.0556 111	0.08809 0.3579 111	0.11239 0.2402 111	-0.08809 0.3579 111	-0.26650 0.0038 116	-0.12239 0.1906 116	-0.20041 0.0310 116	1.00000 116	0.02336 0.8034 116	-0.55154 0.0035 26
tmc Total Moisture Content	-0.10772 0.1063 226	0.01847 0.7562 285	0.35592 <.0001 325	0.31399 <.0001 325	0.23198 <.0001 325	-0.31399 <.0001 325	-0.08352 0.1288 332	-0.10458 0.0668 308	-0.07120 0.2127 308	0.02336 0.8034 116	1.00000 334	-0.18630 0.3332 29
pH pH at the analysis	0.16897 0.4093 26	-0.38215 0.0448 28	-0.12174 0.2790 81	0.00948 0.9330 81	-0.11543 0.5912 24	-0.00948 0.9330 81	-0.20919 0.2761 29	-0.08621 0.6566 29	-0.19614 0.3079 29	-0.55154 0.0035 26	-0.18630 0.3332 29	1.00000 121

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