

# Research Article

# Drip Loss Assessment by Different Analytical Methods and Their Relationships with Pork Quality Classification

Robledo de Almeida Torres Filho,<sup>1</sup> Henrique Pereira Cazedey,<sup>2</sup> Paulo Rogério Fontes,<sup>3</sup> Alcinéia de Lemos Souza Ramos,<sup>2</sup> and Eduardo Mendes Ramos<sup>2</sup>

<sup>1</sup>Instituto de Ciências Exatas e Tecnológicas, Universidade Federal de Viçosa (UFV), Campus UFV Florestal, 35690-000 Florestal, MG, Brazil

<sup>2</sup>Departamento de Ciência dos Alimentos, Universidade Federal de Lavras (UFLA), P.O. Box 3037, 37200-000 Lavras, MG, Brazil <sup>3</sup>Departamento de Tecnologia de Alimentos, Universidade Federal de Viçosa (UFV), 36570-000 Viçosa, MG, Brazil

Correspondence should be addressed to Eduardo Mendes Ramos; emramos@dca.ufla.br

Received 21 July 2016; Revised 23 December 2016; Accepted 5 February 2017; Published 12 March 2017

Academic Editor: Susana Fiszman

Copyright © 2017 Robledo de Almeida Torres Filho et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

We analyzed drip loss in pork by comparing the standard bag (DL), filter-paper wetness (FPW), and EZ-DripLoss methods by weighing the meat juice container and dabbed sample after 24 h and 48 h. Samples were classified into quality categories based on pH, color, and drip loss. The relationship between DL and FPW revealed the cut-off of 5% DL as corresponding to FPW of 139 mg; 1.89% when analyzed by weighing meat juice container or dabbed sample after 24 h; and 3.18% and 3.74% for those analyzed by weighing both meat juice container and dabbed sample after 48 h, respectively. Highest correlations were observed between DL and EZ when the meat juice container was weighed after 48 h (r = 0.86). The EZ-DripLoss method in which the meat juice container was weighed after 48 h (r = 0.86). The EZ-DripLoss method. Therefore, this method is recommended for meat categorization because of its greater standardization and ease of application.

## 1. Introduction

The loss of fluids from pork is important for the industry because of its economic implication. Water accounts for approximately 75% of the weight of meat, and the ability of muscle to retain moisture is key to many meat-quality parameters held in high regard by the industry and consumers [1]. High drip losses lead to losses in terms of appearance, texture, nutritional value, and attractiveness, thereby compromising the quality of fresh meat and its processing [2]. Larger drip losses are usually linked to a greater level of protein denaturation, because the water-holding capacity (WHC) of meat is affected by the state of the muscle proteins. A rapid pH decline postmortem may lead to protein denaturation, with serious consequences for the color, tenderness, and WHC, generating pale, soft, and exudative (PSE) meat [3]. Although the initial pH (measured at 45 min postmortem) may be used as an indicator of PSE condition, its application is limited, because it does not allow for the prediction of all quality

categories [4]. Thus, meat drip loss and lightness form the base of the definition of the pork quality categories, including RSE (reddish-pink, soft, and exudative), PFN (pale, firm, and nonexudative), RFN (reddish-pink, firm, and nonexudative), and DFD (dark, firm, and dry) meat [5].

Because drip loss has stood out as one of the most important parameters of meat-quality evaluation, several methods have been developed to determine it [6, 7]. The percentage drip loss measured by the bag method (DL), proposed by Honikel [8], is recognized internationally as the standard methodology, but it requires a larger space and careful handling of samples. The filter-paper wetness (FPW) method, described by Kauffman et al. [9], is recognized as the simplest and fastest technique to evaluate the meat WHC, and it is reported as being highly correlated with the DL measurements. Later, Rassmussen and Andersson [10] suggested a method involving drip loss containers, referred to as the EZ-DripLoss (EZ) method. This method uses less space and is more easily reproduced and less sensitive to sample handling than the DL method. Furthermore, EZ has also been reported as being highly correlated with DL [2, 11].

However, although all these three methods can measure WHC, they differ in the procedures through which the exudate is measured. The DL and EZ techniques are gravimetric methods in which the meat is left suspended in an airtight container for a long period (24 h to 48 h) to drip [8, 10]; the only force on the meat is gravity. Filterpaper wetness, however, is a gravimetric method based on the amount of fluid on the exposed cut surfaces of muscles absorbed immediately (<3 s) by a filter-paper [9]. According to Honikel and Hamm [13], this absorption method is a rapid alternative to drip loss measurements, but its use in relation to other WHC measurements must be proven. In addition to the differences in the physical principle of water release (gravitation and/or capillarity), the surface area, the weight, the fiber direction of the sample, and the storage time for drip loss determination are also important [3, 13] and can affect the results. The DL method is carried out with cuboid samples of 40-100 g for 48 h storage, whereas the EZ method uses cylindrical samples of 5-10 g measured after 24 h of storage. Moreover, variations in EZ procedures were suggested by Correa et al. [14], namely, use of 48 h storage time and weighing dabbed samples instead of containers, as proposed in the original procedure.

Since the amount of fluids lost affects both qualitative and quantitative aspects of a muscle, as muscle is used for food, the industry requires methods that can easily predict the water-holding capacity (WHC) of the meat product. Because of the variation in the employed methods, results for drip loss in the literature are difficult to compare. For this reason, there have been efforts to create international reference methods to ensure comparability among drip loss measurements [6]. Therefore, the first objective of the present study was to investigate the relationship between the DL method, representing an accepted drip loss measurement, and two other promising measurements: EZ methods (including different sample handling techniques) and FPW. The second objective was to analyze relationships between these WHC measurements and pork quality traits.

### 2. Material and Methods

2.1. Animals, Slaughter, and Carcass Handling Procedures. This study involved 60 pigs, 10 selected randomly per day during six days (commercial cross Large White × Landrace), weighing  $105\pm10$  kg, obtained from different producers from a commercial slaughterhouse, in Lavras, Minas Gerais, Brazil. The animals were electrically stunned and bled in the vertical position by sectioning their jugular veins and carotid arteries, following the standards regulated by the Brazilian legislation. After 45 min postmortem, the initial pH was determined in the Longissimus thoracis (LT) muscle between the 9th and 10th ribs. The carcasses were identified and kept refrigerated  $(1 \pm 1^{\circ}C)$  for 24 h. Afterwards, the LT muscles were removed, packaged, and transferred at 4°C to the Laboratory of Meat and Meat Products Technology (Lab Carnes) at the Federal University of Lavras (UFLA) for analysis of ultimate pH, lightness ( $L^*$ ), and water-holding capacity (WHC).

2.2. Meat-Quality Analysis. The initial pH ( $pH_{45 min}$ ) and ultimate pH ( $pH_{24 h}$ ) of the LT muscle were measured in triplicate 45 min and 24 h postmortem, respectively, using a HI99163 portable meter (Hanna Instruments Brazil, São Paulo, SP, Brazil) with a spear-tipped glass electrode.

Three slices with 2.5 cm thickness were cut after the 10th rib (caudal end) from each loin. Slices were allocated at random for the drip loss methods: a slice was used for filterpaper wetness (FPW) and lightness measurements; another slice was used for the drip loss bag method; and the last slice was used for the EZ-DripLoss method.

Filter-paper wetness was determined in a single replicate, according to the methodology described by Kauffman et al. [9]. The loin chops were exposed to the environment (blooming) at room temperature for 30 min before being analyzed. A qualitative filter-paper (125 mm in diameter, Whatman<sup>®</sup> Grade 1) was weighed, placed on the meat surface for 3 s, and then weighed again. The FPW was expressed as the weight (mg) of the absorbed exudate. Subsequently, the sample lightness ( $L^*$ ) was obtained from the average of three readings taken at different positions on the meat surface using a CM-700 spectrophotometer (Konica Minolta, Inc., Tokyo, Japan) set to 8 mm aperture, specular component excluded (SCE), illuminant  $D_{65}$ , and 10° observer angle.

Drip loss was determined by the standard bag method [8] and by the original [10] and modified [14] EZ-DripLoss methods. In the bag method (DL), drip loss was measured as the weight loss during suspension of a standardized  $(40-50 \text{ g and approximately } 30 \times 60 \times 25 \text{ mm})$  muscle sample (in an airtight container over 48 h at 4°C). Drip loss was expressed as a percentage relative to the initial weight. For EZ-DripLoss (EZ), two samples (~6.4 g each) were taken in dorsal and middle positions from the 2.5 cm thick steaks with a cork borer (25 mm diameter) and placed in a funnel-shaped plastic container (Christensen Aps Industrivaenget, Hillerød, Denmark). Muscle cores  $(EZ_S)$  and containers  $(EZ_C)$  were weighed before and after storage for 24 h ( $EZ_{S24}$  and  $EZ_{C24}$ ) and 48 h (EZ  $_{\rm S48}$  and EZ  $_{\rm C48}$  ) at 4°C. Before each final weighing, the surface of the samples was dabbed gently with paper towels as suggested by Correa et al. [14]. The EZ drip loss was expressed as a percentage relative to the initial weight.

2.3. Pork Classification into Quality Categories. Pork samples were classified according to  $pH_{24h}$ , lightness ( $L^*$ ), and DL parameters into one of the following four quality classes, as defined by Warner et al. [12]:

Pale, soft, and exudative (PSE):  $L^* > 50$ , drip loss > 5%, and pH<sub>24 h</sub> < 6.0;

Reddish-pink, soft, and exudative (RSE):  $L^* = 42-50$ , drip loss > 5%, and pH<sub>24 h</sub> < 6.0;

Reddish-pink, firm, and nonexudative (RFN):  $L^* = 42-50$ , drip loss < 5%, and pH<sub>24 h</sub> < 6.0;

Dark, firm, and dry (DFD):  $L^* < 42$ , drip loss < 5% and pH<sub>24 h</sub> ~ 6.0.

Samples that were not classified into any of these categories were identified as "unclassified" (UC).

Characteristic	Mean	SE	Min	Max	CV (%)
pH <sub>45 min</sub>	5.92	0.03	5.21	6.48	4.37
pH <sub>24h</sub>	5.63	0.02	5.35	6.04	2.79
Lightness $(L^*)$	51.72	0.45	41.63	61.81	6.70
WHC methods					
DL (%)	6.54 <sup>a</sup>	0.29	1.06	11.69	34.87
EZ <sub>C24</sub> (%)	3.10 <sup>d</sup>	0.24	0.13	8.51	58.71
EZ <sub>S24</sub> (%)	3.13 <sup>d</sup>	0.26	0.06	8.36	63.95
EZ <sub>C48</sub> (%)	4.40 <sup>c</sup>	0.27	0.52	9.64	47.89
EZ <sub>S48</sub> (%)	5.19 <sup>b</sup>	0.33	0.39	10.80	49.45
FPW (mg)	174.6	9.9	33.9	372.1	43.95

TABLE 1: Means, standard error (SE), minimum (Min), maximum (Max), and coefficients of variation (CV) for meat quality characteristics measured on pork loin (*Longissimus thoracis* muscle; n = 60).

WHC = water-holding capacity; DL = drip loss by the bag method after 48 h storage;  $EZ_{C24} = EZ$ -DripLoss by weighing containers after 24 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 24 h storage;  $EZ_{C48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{F48} = EZ$ -DripLoss by weighing container

 $^{a-d}$ Means followed by different letters, within the WHC methods, differ (*P* < 0.05) by Tukey's test.

2.4. Statistical Analysis. The statistical analyses were performed on SAS 9.2 (Statistical Analysis System (SAS) Institute Inc., Cary, NC, USA) software at a significance level of 5%. The animal was considered a block, because the analytical methods were measured in each animal. Analysis of variance (ANOVA) and Tukey's test were performed to evaluate the differences between drip loss methods. Pearson's correlation analysis was performed among the quality attributes, whose coefficients (*r*) were tested by Student's *t*-test. Regression analyses of EZ and FPW as a function of DL were performed to determine the equivalent values between these WHC parameters. ANOVA and (when necessary) Tukey's test were conducted to evaluate differences in WHC measured by all analytical methods among the meat-quality categories, classified according to the reference criteria.

#### 3. Results and Discussion

Mean values, standard deviations, and coefficients of variation of pH,  $L^*$ , and water-holding capacity (WHC) obtained with different methodologies are presented in Table 1.

3.1. Differences between Bag Method and EZ-DripLoss. The drip loss by the bag method (DL) was greater (P < 0.05) than that for all EZ methods, with 1.35 and 2.14 percentage points more drips than EZ<sub>S48</sub> (sample was dabbed and weighed) and EZ<sub>C48</sub> (weighing the container with the meat juice), respectively. Christensen [11] also observed a 1.20 percentage points greater DL compared with the EZ method (weighing the container as EZ<sub>C</sub>), both evaluated after 24 h storage. However, Otto et al. [2] reported that DL was 1.64 percentage points lower than the EZ method (evaluated as EZ<sub>C</sub>) after 48 h storage. These authors justified these results as being due to the greater surface area/weight ratio for the EZ method (2.6) compared with the bag method (1.3). The surface area/weight ratio of the samples was postulated to be the reason for differences due to the difference in sample size [11, 13].

However, in the present study, the mean surface area/weight ratio was approximately 4.6 using the EZ method and 1.8 using the DL method; even so, we observed a greater drip loss for the DL method. It is more likely that the surface area in which water primarily escapes is more important.

Postmortem changes in the myofilament lattice spacing generate the driving force for drip loss, but the actual direction of the muscle fibers during storage has a great influence on drip loss [3, 13]. The water is expelled from the myofibrillar structure by lateral and transversal shrinkage of the myofibrils, passes through the sarcoplasmic membrane (which becomes more permeable after rigor) and accumulates in the extracellular space. The separation of fiber bundles and individual muscle cells forms gaps ("drip channels") that guide the water from the extracellular space to the meat surface [1, 3, 15, 16]. Thus, water escapes from the muscle primarily by these drip channels, formed along the length of the muscle fiber. Since the Longissimus muscle fiber angle relative to the steak surface was approximately 45° [17], the surface areas where the water could be lost more easily in the form of drip (by the "drip channels") are the base of the cylindrical samples (EZ method) and the length of the cuboid samples (DL method). This may explain the greater drip losses observed in the DL method, since the length surface area was 3.7 times greater than the base surface area of the EZ samples.

3.2. Differences between EZ-DripLoss Procedures. As expected, samples stored for 48 h had a greater (P < 0.05) drip (2.06 percentage points for EZ<sub>S</sub> and 1.30 percentage points for EZ<sub>C</sub>) than samples stored for 24 h. Correa et al. [14] also observed a lower drip (1.04 percentage points) in samples evaluated after 24 h as compared with those evaluated after 48 h storage. Fluid losses increase with time for several days, because exudation is a slow process [13]. The water must be expelled from the myofibrillar lattice to accumulate in the extracellular space, being progressively drained (through the drip channels) out of the muscle as purge [1].

	$pH_{45min}$	$pH_{24h}$	$L^*$	DL (%)	FPW (mg)	EZ <sub>C24</sub> (%)	EZ <sub>S24</sub> (%)	EZ <sub>C48</sub> (%)
pH <sub>24 h</sub>	-0.04							
Lightness $(L^*)$	-0.21	$-0.34^{**}$						
DL (%)	$-0.52^{**}$	-0.19	0.62**					
FPW (mg)	$-0.53^{**}$	-0.14	0.53**	0.67**				
EZ <sub>C24</sub> (%)	$-0.48^{**}$	0.10	$0.47^{**}$	0.83**	0.59**			
EZ <sub>S24</sub> (%)	$-0.48^{**}$	0.11	$0.49^{**}$	0.83**	$0.54^{**}$	0.97**		
EZ <sub>C48</sub> (%)	$-0.40^{**}$	0.05	0.52**	0.86**	0.58**	0.97**	0.96**	
EZ <sub>S48</sub> (%)	$-0.43^{**}$	0.05	0.55**	$0.84^{**}$	$0.54^{**}$	0.93**	0.96**	0.95**

TABLE 2: Correlation coefficients among the pork loin (*Longissimus thoracis* muscle; n = 60) quality attributes.

DL = drip loss by the bag method after 48 h storage;  $EZ_{C24} = EZ$ -DripLoss by weighing containers after 24 h storage;  $EZ_{S24} = EZ$ -DripLoss by weighing dabbed samples after 24 h storage;  $EZ_{C48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage; and FPW = filter-paper wetness (mg). \* P < 0.05. \*\* P < 0.01.

At 48 h storage, the average EZ drip loss value in dabbed samples (EZ<sub>548</sub>) was higher by 0.79 percentage points (P <0.05) than in nondabbed samples (weighing the container; EZ<sub>C48</sub>). Correa et al. [14] also observed a greater drip loss (1.80 percentage points) when dabbed samples were weighed in relation to the weight of container after 24 and 48 h storage. According to these authors, the dabbing procedure helped to remove the exudate still present on the core surface at the end of storage time to allow for the determination of the real water lost by the muscle core. However, in the present experiment, the EZ drip losses measured after 24 h did not differ (P >0.05) among each other. This may be due to differences in water loss rate between studies. In the experiment of Correa et al. [14], about 80% (85% for dabbed sample and 76% for nondabbed sample) of the EZ drip loss occurred in the first 24 h, while, in the present experiment, only 65% (60% for dabbed sample and 70% for nondabbed sample) of water had dripped in this storage time. Addressing the DL method, Otto et al. [2] reported that only 58% of the drip loss occurred in the first 24 h of the 48 h under evaluation.

Christensen [11] stated that absence of superficial dabbing helps to reduce the influence of the handler in the analysis. According to Correa et al. [14], this affects the measurement reliability, because the weight of the container with exudate does not take into account any remaining drip on the sample surface, leading to an underestimated value of the drip exuded from the pork sample during storage. However, in our results, this was true only for drip loss measured after 48 h storage. Moreover, drip loss evaluation after 24 h may underestimate fluid loss during the pork storage. For this reason, Kauffman et al. [18] and Honikel [8] recommend a storage time of 48 h to evaluate the drip loss, regardless of the adopted method.

All the coefficients of variation of EZ methods were greater (48 to 64%) than that of DL (35%) (Table 1). Nevertheless, this larger variation may be interesting to facilitate the discrimination of the samples into quality categories, given that the samples showed a wide variability, which can be observed by the ranges of  $pH_{24h}$  (5.35 to 6.04) and lightness (41.6 to 61.8). As observed by Correa et al. [14], the

coefficients of variation for dabbed samples were higher than for nondabbed samples. These authors reported coefficients between 39 and 52% for EZ methods, whereas Otto et al. [2] observed a coefficient of variation of 48% for both DL and  $\rm EZ_{C48}$ .

3.3. Correlations between WHC and Other Evaluated Meat Characteristics. The EZ drip loss was highly and positively correlated (r > 0.83; P < 0.05) with DL (Table 2). Christensen [11] observed a high correlation (r = 0.85) between 24 h DL and EZ<sub>C24</sub>, whereas Otto et al. [2] reported a high correlation (r = 0.86) between EZ<sub>C48</sub> and DL evaluated after both 24 h and 48 h. The EZ drip loss also showed a high positive correlation (r > 0.93; P < 0.05) with each other, indicating that all are satisfactory to obtain similar results. On the other hand, the EZ drip losses showed an intermediate positive correlation (r = 0.54 to 0.59; P < 0.05) with FPW, which presented intermediate positive correlation (r > 0.67; P < 0.05) with DL. This differs from the high correlation coefficient (r > 0.96) reported by Kauffman et al. [9] between FPW and DL.

All the WHC methods showed intermediate negative correlations (r = -0.53 to -0.40; P < 0.05) with pH<sub>45 min</sub>. This is coherent, because the rate of pH decline affects the protein denaturation, the myofilament lattice spacing, and the shrinking of the muscle fiber, which makes it essential for the determination of meat-quality. A high rate of postmortem pH fall may have negative consequences for color, tenderness, and WHC, generating PSE. Considering that 85% of the water in the muscle is located between the myofibrils, sustained by capillary forces, the denaturation of these proteins or sarcomere shortening causes a reduction in WHC, thus increasing the water loss from the meat [1, 3]. Correa et al. (2007) also observed similar correlations (-0.59 to -0.48)between the EZ methods and pH<sub>45 min</sub>, and Otto et al. [2] reported negative intermediate correlations (-0.52 to -0.48)of DL and  $\mathrm{EZ}_{\mathrm{C48}}$  with  $\mathrm{pH}_{\mathrm{45\,min}}.$ 

The correlations of pH<sub>45 min</sub> with EZ drip losses evaluated after 24 h (r = -0.48; P < 0.05) were slightly stronger as compared with those evaluated after 48 h (r = -0.43 to -0.40;

	Quality class						
Characteristic	PSE	RSE	RFN	DFD			
Ν	36	8	7	1			
pH <sub>45 min</sub>	$6.04 \pm 0.05^{a}$	$5.99 \pm 0.13^{a}$	$6.26 \pm 0.04^{a}$	6.44 <sup>a</sup>			
pH <sub>24h</sub>	$5.60 \pm 0.02^{\rm b}$	$5.65 \pm 0.05^{\rm b}$	$5.67 \pm 0.04^{\rm b}$	6.04 <sup>a</sup>			
Lightness $(L^*)$	$53.52 \pm 0.42^{a}$	$48.24 \pm 0.61^{b}$	$47.85 \pm 0.47^{b}$	41.63 <sup>c</sup>			
WHC method							
DL (%)	$7.81 \pm 0.27^{a}$	$6.54\pm0.32^{ab}$	$3.68 \pm 0.51^{\rm bc}$	1.06 <sup>c</sup>			
EZ <sub>C24</sub> (%)	$3.86 \pm 0.29^{a}$	$3.22 \pm 0.50^{a}$	$1.27 \pm 0.26^{a}$	0.51 <sup>a</sup>			
EZ <sub>S24</sub> (%)	$4.00 \pm 0.30^{a}$	$3.21 \pm 0.65^{a}$	$1.09 \pm 0.22^{a}$	0.36 <sup>a</sup>			
EZ <sub>C48</sub> (%)	$5.38 \pm 0.29^{a}$	$4.54\pm0.52^{\rm ab}$	$1.82 \pm 0.36^{\rm bc}$	0.75 <sup>c</sup>			
EZ <sub>S48</sub> (%)	$6.40 \pm 0.35^{a}$	$5.39 \pm 0.73^{a}$	$2.17 \pm 0.51^{ab}$	$0.42^{b}$			
FPW (mg)	$211.6 \pm 11.5^{a}$	$139.3 \pm 17.4^{ab}$	$100.6 \pm 17.9^{ab}$	33.9 <sup>b</sup>			

TABLE 3: Meat quality characteristics (mean  $\pm$  standard error) of four quality groups by Warner et al. [12] criteria of pork loin (*Longissimus thoracis* muscle; n = 52)<sup>1</sup> samples.

PSE = pale, soft, and exudative; RSE = reddish-pink, soft, and exudative; RFN = reddish-pink, firm, and nonexudative; DFD = dark, firm, and dry; N = number of samples assigned to each pork quality class; WHC = water-holding capacity;  $DL = drip loss by the bag method after 48 h storage; <math>EZ_{C24} = EZ$ -DripLoss by weighing containers after 24 h storage;  $EZ_{S24} = EZ$ -DripLoss by weighing dabbed samples after 24 h storage;  $EZ_{C48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss

 $^{1}$ Eight samples (from 60) could not be classified into any pork quality category, and so they were excluded from the analysis.

<sup>a-c</sup>Means followed by different letters in the row differ (P < 0.05) by Tukey's test.

P < 0.05). A possible explanation for this is that PSE, which were in a larger proportion in the present study, have most of their fluid dripping out on the first day postmortem, whereas the drip in RFN is released more slowly [1, 3, 14]. Correa et al. [14], however, observed a lower correlation of pH<sub>45 min</sub> with EZ<sub>C24</sub> (r = -0.48) than with the other EZ methods (-0.59 to -0.57).

Although drip loss or purge loss are generally well correlated with ultimate pH, none of the evaluated analytical methods of WHC was correlated (P > 0.05) with pH<sub>24h</sub>. Correa et al. [14] observed low negative correlations (-0.19 to -0.28) between EZ methods and pH<sub>24h</sub>, while Otto et al. [2] reported low negative correlations of DL (r = -0.37) and EZ<sub>C48</sub> (r = -0.36) with pH<sub>24h</sub>. The absence or lower correlation coefficients of drip loss with pH<sub>24h</sub> as compared with pH<sub>45 min</sub> may be due to its smaller variation (Table 1) and to the greater proportion of PSE in the present study, as explained previously.

All the WHC analytical methods showed intermediate positive correlations (r = 0.47 to 0.62; P < 0.05) with lightness ( $L^*$ ), despite the absence (P > 0.05) of correlations between  $L^*$  and  $pH_{45 \min}$  and the low negative correlations (r = -0.34; P < 0.05) of  $L^*$  with pH<sub>24h</sub>. Together with the pH fall, the denaturation of myofibrillar and sarcoplasmic (myoglobin) proteins and the expulsion of the water from the myofibrils towards the extracellular space during rigor mortis may lead to structural changes that increase light scattering [1, 3, 19], making the meat paler (greater  $L^*$ ). However, a high correlation between pH and color has been reported when a wide color range was evaluated, that is, a greater proportion of both extreme conditions of PSE and DFD quality [19]. In the present study, the greater correlation coefficients between drip losses and  $L^*$  may be explained by the greater proportion of PSE, which are paler and more exudative simultaneously. Correa et al. [14] also observed

intermediate positive correlations (0.48 to 0.51) between EZ methods and  $L^*$ , whereas Otto et al. [2] reported low correlations of DL (r = 0.38) and EZ<sub>C48</sub> (r = 0.42) with  $L^*$ .

3.4. Pork Classification into Quality Categories according to WHC. Several criteria have been proposed [12, 20–24] to classify pork into different quality categories, but there is no international consensus on what criteria should be used. The classification ability depends on quality attributes used in the characterization of pork, which could explain the wide variation in the incidence of PSE reported in the literature [25]. The use of distinct criteria established for different attributes can be explained by the fact that quality is defined according to several perspectives of interest [4]. In this regard, lightness and WHC were key attributes, since they allow for the separation of meat by its color appearance, which directly affects consumer acceptance, and, by its exudation, which affects yield for the industry.

To determine the effect of the WHC measurement technique on the categorization of pork into quality classes, we use the criteria of Warner et al. [12] (Table 3). At the time of statistical analysis, eight (13%) samples could not be classified into any pork quality category, and so they were excluded from the analysis. The DL results are quite similar to those reported by Kauffman et al. [20] and van Laack et al. [26], who observed a mean percentage drip loss of 7% for the exudative meats (PSE and RSE), 3% for RFN, and 2% for DFD meats. As reported by these authors, there were no differences (P > 0.05) between drip losses of PSE and RSE samples. Joo et al. [24] reported a mean drip loss of 10.4% for PSE samples versus 7.4% for RSE samples, but this difference may have been exaggerated, since they selected extreme pale and exudative samples. For FPW, however, the fluid weight values observed in this experiment were much larger than those described by Kauffman et al. [20] and Joo et al. [24]. These

Quality class	DL (%)	EZ <sub>C24</sub> (%)	EZ <sub>S24</sub> (%)	EZ <sub>C48</sub> (%)	EZ <sub>S48</sub> (%)	FPW (mg)
PSE	60	60	58	63	55	58
RSE	13	13	8	12	10	10
RFN	12	12	17	13	15	15
DFD	2	2	2	2	2	2
Unclassified	13	13	15	10	18	15

TABLE 4: Frequency (%) of occurrence of each quality class by Warner et al. [12] criteria following characterization by the WHC method measured on pork loin (*Longissimus thoracis* muscle; n = 60) samples.

PSE = pale, soft, and exudative; RSE = reddish-pink, soft, and exudative; RFN = reddish-pink, firm, and nonexudative; DFD = dark, firm, and dry; DL = drip loss by the bag method after 48 h storage;  $EZ_{C24} = EZ$ -DripLoss by weighing containers after 24 h storage;  $EZ_{S24} = EZ$ -DripLoss by weighing dabbed samples after 24 h storage;  $EZ_{C48} = EZ$ -DripLoss by weighing containers after 48 h storage;  $EZ_{S48} = EZ$ -DripLoss by weighing dabbed samples after 48 h storage; and FPW = filter-paper wetness (mg). EZ and FPW criteria used were equivalent to 5% DL.

authors observed, for example, an average fluid weight of 114, 104, and 147 mg, respectively, for FPW of PSE meat group, which corresponds to the values observed for RFN and RSE meats in this experiment. This discrepancy can be explained by the fact that we used a filter-paper with 125 mm diameter to conduct the FPW test, while Kauffman et al. [9] described a filter-paper with 55 mm diameter. Therefore, the high values of FPW were due to the greater absorption of fluid into the filter-paper, since it had a larger contact area with the meat surface, covering most of the meat surface.

Regarding the differences in exudation loss among the pork quality categories, only  $EZ_{C48}$  attained the same level of distinction as the standard DL method (Table 3). This was probably a result of the greater mean values of  $EZ_{C48}$  in relation to the EZ drip losses evaluated after 24 h and its lower dispersion (standard deviation) as compared with  $EZ_{S48}$ . In addition,  $EZ_{C48}$  had a higher (P < 0.05) correlation coefficient with DL than the other drip tests (Table 2). When the 24 h EZ-DripLoss methods were used, the drip values did not differ between any of the pork quality classes, despite the high (r = 0.83; P < 0.05) correlation with DL.

The bag method (DL) was used by some authors [12, 20, 24] to assess drip loss, with 5-6% DL arbitrarily suggested as the standard value to separate acceptable from unacceptable WHC to categorize pork into quality classes. Other authors [21, 23] preferred to use the FPW method, with the fluid weight (80–100 mg) used as a criterion for unacceptably exudative meat calculated to correspond to a DL value near 5%. The equation, provided by Kauffman et al. [9], which estimates the fluid weight in FPW, equivalent to the 48 h drip loss by the bag method, is commonly used.

To our knowledge, no studies have suggested the use of EZ drip loss values as a criterion for pork classification into quality categories, although Correa et al. [14] used this method to assess drip loss and to classify pork meats with criterion values established for DL. It must be stressed that those authors utilized the criterion of 5% drip loss by the DL method for the classification of pork quality by the EZ method, without, however, considering the large differences in the absolute values that exist between these methods.

Therefore, to use FPW or EZ values as WHC parameters in the classification of pork into quality categories, it is first necessary to determine the values equivalent to 5% DL used by Warner et al. [12]. In this experiment, the Kauffman et



FIGURE 1: Linear regression of filter-paper wetness (FPW) as a function of 48 h drip loss by the bag method (DL) in pork loins (n = 60). Fluid weight (FPW), mg =  $26.06 + 22.69 \times DL \pm 57.1$  mg (SE),  $R^2 = 0.46$ , and P < 0.0001.

al. [9] equation does not fit, since we used a filter-paper of larger diameter, which absorbs more fluid. Evaluating the relationship between DL and FPW in the loin muscles (Figure 1), the cut-off of 5% DL corresponds to FPW value of 139 mg. However, parts of the filter-paper may not get in contact with the meat of the loin from smaller animals, and thus they may provide a different water absorption performance. Similarly, the evaluation of the relationship between DL and EZ methods (Figure 2) indicated that 5% DL corresponds to 1.89%  $EZ_{24}$  ( $EZ_{C24} + EZ_{S24}$ ), 3.18% $EZ_{C48}$ , and 3.74% $EZ_{S48}$ .

Using approximate values of 5% DL equivalents (2% EZ<sub>24</sub>, 3%EZ<sub>C48</sub>, and 4%EZ<sub>S48</sub>), together with pH<sub>24h</sub> and *L*<sup>\*</sup> criteria of Warner et al. [12], samples were distributed according to the WHC method (Table 4). In this way, the difference between WHC procedures in the rate and extent of exudation measurements was the only source of shifting from one class to another. Overall, in the EZ method, weighing the sample instead of the container (EZ<sub>S24</sub> and EZ<sub>S48</sub>) allowed the detection of a slightly greater percentage of RFN and unclassified samples over the percentage of meat classified as exudative (RSE and PSE). The same effect was observed when classification was performed using the FPW method as a predictor of WHC. The conventional EZ method (24 h storage and weighing the container; EZ<sub>C24</sub>) allowed for a



FIGURE 2: Regressions of 24 h (EZ<sub>24</sub>) and 48 h, dabbed (EZ<sub>C48</sub>) and nondabbed (EZ<sub>S48</sub>) EZ-DripLoss as a function of 48 h drip loss by the bag method (DL) in pork loins (n = 60). EZ<sub>24</sub>, % = 0.33 + 0.07 × DL + 0.05 × DL<sup>2</sup> ± 1.02% (SE),  $R^2 = 0.72$ , and P < 0.0001; EZ<sub>C48</sub>, % = -0.78 + 0.79 × DL ± 1.10% (SE),  $R^2 = 0.74$ , and P < 0.0001; and EZ<sub>S48</sub>, % = -0.98 + 0.94 × DL ± 1.41% (SE),  $R^2 = 0.70$ , and P < 0.0001.

classification of samples identically to that by 48 h DL, while weighing the EZ container after 48 h storage ( $EZ_{C48}$ ) slightly increased the percentage of PSE meat with a slight reduction of the percentage of unclassified meat.

#### 4. Conclusion

The different WHC prediction methods provide different drip values in absolute terms. Therefore, each WHC method needs a corresponding threshold value to be used to classify pork into different quality classes. The relationship between drip loss by the bag (DL) and filter-paper wetness (FPW) methods indicated that the cut-off of 5% DL corresponds to FPW value of 139 mg. Similarly, the relationship between the DL and EZ-DripLoss (EZ) methods revealed 5% DL as corresponding to 1.89% when analyzed by weighing meat juice container or dabbed sample after 24 h and 3.18% and 3.74% by weighing both meat juice container and dabbed sample after 48 h, respectively.

Sample dabbing did not improve the reliability of the EZ-DripLoss methodology for the drip loss assessment and overall pork quality evaluation, but extending the conventional storage time from 24 h to 48 h is recommended to increase the accuracy of the method. However, the EZ method by weighing the meat juice container after 24 h was able to distinguish drip loss into meat-quality categories, by the bag method. Therefore, this method is recommended for meat categorization, given its greater standardization and ease of application.

#### **Additional Points**

*Practical Applications.* Because of the variation in employed methods, the results for drip loss and classification into different meat-quality categories in the literature are difficult to compare. The present study reports the relationship

between drip loss by the standard bag method and other promising measurements—EZ methods, by different sample handling techniques, and FPW—to allow this comparison in the literature. The analysis of relationships between these WHC measurements and pork quality traits indicated a corresponding threshold value to be used to classify pork into different quality classes.

#### **Conflicts of Interest**

The authors declare that there are no conflicts of interest regarding the publication of this paper.

#### Acknowledgments

This study was financed by the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq; Grant no. 476355/2012-5); the authors gratefully acknowledge their assistance. The authors would also like to thank the Fundação de Amparo à Pesquisa do Estado de Minas Gerais (FAPEMIG), the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), and CNPq for the fellowship granted to the second (PIBIC/CNPq) and third (postdoctoral PMPD II, CAPES/FAPEMIG) authors.

#### References

- E. Huff-Lonergan and S. M. Lonergan, "Mechanisms of waterholding capacity of meat: the role of postmortem biochemical and structural changes," *Meat Science*, vol. 71, no. 1, pp. 194–204, 2005.
- [2] G. Otto, R. Roehe, H. Looft, L. Thoelking, and E. Kalm, "Comparison of different methods for determination of drip loss and their relationships to meat quality and carcass characteristics in pigs," *Meat Science*, vol. 68, no. 3, pp. 401–409, 2004.

- [3] J. M. Hughes, S. K. Oiseth, P. P. Purslow, and R. D. Warner, "A structural approach to understanding the interactions between colour, water-holding capacity and tenderness," *Meat Science*, vol. 98, no. 3, pp. 520–532, 2014.
- [4] E. M. Ramos and L. A. M. Gomide, Avaliação da Qualidade de Carnes: Fundamentos e Metodologias, UFV, Viçosa, Brazil, 2007.
- [5] R. L. J. M. Van Laack and R. G. Kauffman, "Glycolytic potential of red, soft, exudative pork longissimus muscle," *Journal of Animal Science*, vol. 77, no. 11, pp. 2971–2973, 1999.
- [6] K. O. Honikel, "Reference methods for the assessment of physical characteristics of meat," *Meat Science*, vol. 49, no. 4, pp. 447–457, 1998.
- [7] C. Kapper, C. J. Walukonis, T. L. Scheffler et al., "Moisture absorption early postmortem predicts ultimate drip loss in fresh pork," *Meat Science*, vol. 96, no. 2, pp. 971–976, 2014.
- [8] K. O. Honikel, "Water binding capacity of meat," *Fleis-chwirtschaft*, vol. 67, pp. 418–428, 1987.
- [9] R. G. Kauffman, G. Eikelenboom, P. G. van der Wal, G. Merkus, and M. Zaar, "The use of filter paper to estimate drip loss of porcine musculature," *Meat Science*, vol. 18, no. 3, pp. 191–200, 1986.
- [10] A. J. Rassmussen and M. Andersson, "New method for determination of drip loss in pork muscles," in *Proceedings of the in 42nd International Congress of Meat Science and Technology (ICoMST* '96), pp. 286–287, Lillehammer, Norway, 1996.
- [11] L. B. Christensen, "Drip loss sampling in porcine m. longissimus dorsi," *Meat Science*, vol. 63, no. 4, pp. 469–477, 2003.
- [12] R. D. Warner, R. G. Kauffman, and M. L. Greaser, "Muscle protein changes post mortem in relation to pork quality traits," *Meat Science*, vol. 45, no. 3, pp. 339–352, 1997.
- [13] K. O. Honikel and R. Hamm, "Measurement of water-holding capacity and juiceness," in *Quality Attributes and Their Measurement in Meat, Poultry and Fish Products*, A. M. Pearson and T. R. Dutson, Eds., pp. 125–161, Blackie Academic & Professional, New York, NY, USA, 1994.
- [14] J. A. Correa, S. Méthot, and L. Faucitano, "A modified meat juice container (EZ-DripLoss) procedure for a more reliable assessment of Drip Loss and related quality changes in pork meat," *Journal of Muscle Foods*, vol. 18, no. 1, pp. 67–77, 2007.
- [15] A. Schäfer, K. Rosenvold, P. P. Purslow, H. J. Andersen, and P. Henckel, "Physiological and structural events post mortem of importance for drip loss in pork," *Meat Science*, vol. 61, no. 4, pp. 355–366, 2002.
- [16] G. Offer and T. Cousins, "The mechanism of drip production: formation of two compartments of extracellular space in muscle Post mortem," *Journal of the Science of Food and Agriculture*, vol. 58, no. 1, pp. 107–116, 1992.
- [17] S. D. Shackelford and T. L. Wheeler, "Slice shear force," in *BEEF FACTS-product enhancement research*, Cattlemen's Beef Board and National Cattlemen's Beef Association, 2009.
- [18] R. G. Kauffman, G. Eikelenboom, P. G. van der Wal, B. Engel, and M. Zaar, "A comparison of methods to estimate waterholding capacity in post-rigor porcine muscle," *Meat Science*, vol. 18, no. 4, pp. 307–322, 1986.
- [19] H. J. Swatland, "Progress in understanding the paleness of meat with a low pH," *South African Journal of Animal Sciences*, vol. 34, no. 6, pp. 1–7, 2004.
- [20] R. G. Kauffman, W. Sybesma, F. J. M. Smulders et al., "The effectiveness of examining early post-mortem musculature to predict ultimate pork quality," *Meat Science*, vol. 34, no. 3, pp. 283–300, 1993.

- [21] R. D. Warner, R. G. Kauffman, and R. L. Russel, "Quality attributes of major porcine muscles: a comparison with the Longissimus Lumborum," *Meat Science*, vol. 33, no. 3, pp. 359– 372, 1993.
- [22] M. B. Maganhini, B. Mariano, A. L. Soares, P. D. Guarnieri, M. Shimokomaki, and E. I. Ida, "Carnes PSE (Pale, Soft, Exudative) e DFD (Dark, Firm, Dry) em lombo suíno numa linha de abate industrial," *Ciencia e Tecnologia de Alimentos*, vol. 27, no. 1, pp. 69–72, 2007.
- [23] L. Faucitano, M. C. Ielo, C. Ster, D. P. Lo Fiego, S. Methot, and L. Saucier, "Shelf life of pork from five different quality classes," *Meat Science*, vol. 84, no. 3, pp. 466–469, 2010.
- [24] S. T. Joo, R. G. Kauffman, B. C. Kim, and C. J. Kim, "The relationship between color and water-holding capacity in postrigor porcine longissimus muscle," *Journal of Muscle Foods*, vol. 6, no. 3, pp. 211–226, 1995.
- [25] H. P. Cazedey, R. d. Torres Filho, P. R. Fontes, A. d. Ramos, and E. M. Ramos, "Comparison of different criteria used to categorize technological quality of pork," *Ciência Rural*, vol. 46, no. 12, pp. 2241–2248, 2016.
- [26] R. L. J. M. van Laack, R. G. Kauffman, W. Sybesma, F. J. M. Smulders, G. Eikelenboom, and J. C. Pinheiro, "Is colour brightness (L-value) a reliable indicator of water-holding capacity in porcine muscle?" *Meat Science*, vol. 38, no. 2, pp. 193–201, 1994.



BioMed Research International







International Journal of Genomics







Submit your manuscripts at https://www.hindawi.com





The Scientific World Journal



Genetics Research International

Archaea



Anatomy Research International





International Journal of Microbiology

International Journal of Evolutionary Biology





Molecular Biology International



Advances in Bioinformatics



Journal of Marine Biology