Evaluation Of Physico-Chemical And Microbiological Quality Of Sheep Meat In The Central Highlands Of Mexico

Araujo-Guzmán, A.F.[1]; *Gutiérrez–Ibáñez, A.T.[1]; Mariezcurrena-Berasain, M.D.[1]; Mariezcurrena-Berasain, M.A.[2]; Pinzón-Martínez, D.L.[1]; Bernal-Martínez, L.R[1]; Salem, A.Z.M.[2]

[1.] Faculty of Agricultural Science, Autonomous University of the State of Mexico
[2] Faculty of Veterinary Medicine and Animal Science, Autonomous University of the State of Mexico
*e-mail address: atarini@uaemex.mx

Abstract: In order to develop a microbiological starting point for sheep meat production from Central Highlands of Mexico, the aim of the current research was to evaluate their microbiological and physico-chemical quality from slaughterhouses. Carcasses, utensils and workers' hands samples were taken according to NOM 092-SSA1-1994, Official Chilean Standard NCh-ISO 17025 of 2005, NF V08-060 Microbiology of food and animal feedings stuffs and European Union Council and Parliament Regulations. The microbiological quality was analyzed by Mesophilic Aerobic, Total Coliforms and Fecal Coliforms values. Most of the slaughterhouses were close to the Superior Limit Permitted (MLP) for Mesophilic Aerobic values (3, 59 $\log_{10}$ UFC/mL) on sheep carcasses. Even though there were no fecal coliforms found on sheep carcasses, they have been found on knives, tables and workers' hands in most of the slaughterhouses. In the physico-chemical variables initial and ultimate pH (45 min and 24 h), initial and ultimate temperature (45 min and 24 h), lightness (L*), redness (a*), yellowness (b*) and tenderness were evaluated and there were significant differences among variables except for initial Temperature ($p \leq 0.05$). In conclusion, although the physico-chemical characteristics were acceptable the Microbiological values overpass the MLP for Mesophilic Aerobic and Fecal Coliform haven't been found any correlation between both of them at the analyst moment. However, because of the high sheep meat production in the Central Highlands of Mexico and these microbiological characteristics it has been concluded that is required a shared responsibility among the productive sector and from the government in order to regulate that production.
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1. Introduction

Sheep in Mexico is oriented to the production of meat and is mainly distributed in the states of Mexico and Hidalgo. The most frequent breeds are: Suffolk, Charollais, Rambouillet, Texel, Pelibuey and Dorper. The most important characteristics of meat are: juiciness, tenderness and flavor, they give more weight to the quality and impact in the decision to purchase (Cuellar, 2007).

In Mexico the consumption of this food is by the traditional culinary process called *Barbacoa* and recently fins cuts. This is the main activity of the Municipalities belonging to the Central Highlands of the State of Mexico. Being the main gatherer and seller of this meat the Municipality of Capulhuac with 1,968 slaughtered heads annually and 685 t of meat monthly (Arteaga, 2011; SIAP, 2012).

The quality factors that have influence on this product include ante mortem (breed, feeding, gender, fasting, transportation, loading and unloading) and *post-mortem* periods (stunning, dressing, chilling and cutting). Therefore, if these factors are not adequately performed quality is affected. In fact, sanitary quality of meat is the additional contributions during dressing, storage, transportation and distribution (Alarcon *et al*., 2006).

The lack of sanitary conditions at the slaughterhouse contributes to microbiological contamination, because the meat offers a high nutrimental environment, which generates pathologies for the consumer. In spite of the few information on these cattle physico-chemical and health characteristics is very important the knowledge within the Central Highlands because of its great marketing (FAO, 2007; Signorini *et al*., 2006).

Therefore, the aim of the present investigation was to evaluate the physico-chemical and microbiological characteristics of sheep meat from slaughterhouses at the Central Highlands of the State of Mexico.

2. Material and Methods

The study was conducted for six months, from August, 2013 to February, 2014. Six inspections were made per abattoir every fifteen days in the

Municipality of Capulhuac, State of Mexico. The physicochemical analyses were carried out in the Laboratories of Veterinarian Medicine and Animal Husbandry Faculty, UAEMex and UNAM Topilejo Unit. The microbiological analyses were carried out in the Laboratory of Agricultural Product Quality, Faculty of Agricultural Sciences, UAEMex.

The study was available only on three of the eight slaughterhouses of the Municipality whose production volume was 420 t monthly. Carcasses, knives, tables and workers' hands samples were taken according to Standard NCh-ISO 17025 Official, 2005 and European Union Council and Parliament Regulations.

1.1 Physico-chemical meat characteristics

After slaughtering, pH 45 min and pH 24 h were measured between the 12th and 13th rib of the carcasses with a potentiometer HANNA HI99163, in agreement to AOAC methodology (1990). Objective color was determined using a Minolta Chroma Meter CR-400 (Honikel, 1998). Finally, tenderness was made under the measure of effort or strength to shear by Warner-Bratzler in the muscle *Longissimus dorsi* according to AMSA method (1992).

1.2 Study of microbiological characteristics

1.2.1 Evaluation of carcass sanitary characteristics

For microbiological analyses a non-destructive method was used in agreement with Standard NCh-ISO 17025 Official, 2005 and European Union Council and Parliament Regulations. Microbiological inocules were taken from four points of the carcass: flank, thorax lateral, brisket and breast (swabbing area 100 cm^2). Samples were prepared under the Official Mexican Standard NOM-110-SSA1-1994 Goods and Services "Preparation and dilution of food samples for microbiological analysis". Mesophilic Aerobic Bacteria, Total and Fecal Coliforms counts were made according to Standards NOM-092-SSA1-1994 (Standard Methods Agar BIOXON), NOM-113-SSA1-1994 and NF V08-60 (1996) from the Association Française de Normalisation (AFNOR), respectively. For the last two, Violet Red Bile Agar (BIOXON) was used.

1.2.2 Evaluation of slaughterhouse sanitary characteristics

Hands (swabbing area 20 cm^2), tables (swabbing area 10 cm^2), knives (both faces and edges) were sampled up at the time of carcasses sampling according t 2005. Microbiological quantifications for Mesophilic Aerobic, Total and Fecal Coliforms were made according to the mentioned standards.

1.3 Experimental design

A completely random design was performed including a variance analysis ($p \leq 0.05$) for abattoirs. Significant differences were compared using a DMS test ($p \leq 0.05$) with the software Stat Graphics Plus 5.0 (1999–2000). The analyzed microorganisms' data (Mesophilic Aerobic, Total and Fecal Coliforms) were reported in $\log_{10}$ CFU/mL. The Mesophilic Aerobic microorganisms were compared with European Union Council and Parliament Regulations Maximum Permissible Limits (MPL), which indicates the MPL (≤ 3.5 $\log_{10}$ CFU/mL). Total Coliforms Standard has not been taken into account, nonetheless for this study was considered as quality indicator. Fecal Coliforms samples should not exceed 1.5 $\log_{10}$ CFU/mL.

3. Results

2.1. Meat physico-chemical characteritics

The data in Table 1 showed pH 45 min, pH 24 h, T 45 min, T 24 h, color with the values of lightness (L*), redness (a*) and yellowness (b*) and tenderness variables.

Regarding to pH 24 h significant differences among slaughterhouses were found, the stated rank for this parameter is from 5,5 to 5,7. Abattoirs 1 and 3 generated the highest values. Concerning to temperature, slaughterhouse 3 is within the normal ranks, 15 °C. In lightness (L*) significant differences among abattoirs were found. In addition, the highest redness (a*) and yellowness (b*) values were found in abattoir 1 and 3, respectively. Moreover, the last one got the highest shear force value (tenderness).

Table 1 Physico-chemical Characteristics of sheep meat

Variable	Abattoir 1(μ ± SD)	Abattoir 2(μ ± SD)	Abattoir 3(μ ± SD)	*P* – value
pH 45	6.17 ± 0.24^{b}	6.02 ± 0.16^{a}	6.27 ± 0.35^{c}	0.0052
pH 24	5.95 ± 0.21^{c}	5.70 ± 0.31^{a}	5.89 ± 0.26^{b}	0.0000
Temperature 45 min (°C)	19.03 ± 1.15	18.46 ± 2.28	19.16 ± 2.73	0.3503NS
Temperature 24 h (°C)	17.02 ± 1.47^{c}	15.11 ± 2.55^{a}	16.31 ± 1.38^{b}	0.0000
L*	39.34 ± 7.08^{c}	34.22 ± 5.27^{b}	31.61 ± 4.88^{a}	0.0000
a*	14.05 ± 2.49^{b}	12.30 ± 2.14^{a}	11.54 ± 3.37^{a}	0.0000
b*	6.39 ± 1.47^{a}	8.74 ± 1.56^{c}	7.31 ± 1.65^{b}	0.0000
Tenderness	3.01 ± 0.76^{a}	3.62 ± 0.97^{a}	4.93 ± 1.75^{b}	0.0002

NS= Not Significant; SD= Standard Desviation; μ= Arithmetic Mean; $p \leq 0,05$

2.2. Meat microbiological characteristics

Mesophilic Aerobic values for carcasses, knives and tables have been found significant differences, where standing out abattoir 3 presented the highest values overflowing the MLP. Workers' hands variable had no significant differences (Table 2).

Table 2. Mesophilic Aerobic Microorganisms, for carcasses, knives, workers' hands and tables variables

Mesophilic Aerobic (log_{10} CFU / mL ± SD)	Abattoir 1	Abattoir 2	Abattoir 3	*P* – value
Carcasses	3,40 ± 0,20[a]	3,55 ± 0,30[b]	3,63 ± 0,26[c]	0,0000
Knives	3,26 ± 0,14[a]	3,54 ± 0.21[b]	3,88 ± 0,36[c]	0,0000
Workers' hands	3,50 ± 0,05	3,55 ± 0,18	3,64 ± 0,22	0,1986NS
Tables	3,49 ± 0,17[a]	3,58 ± 0,14[b]	3,71 ± 0,27[c]	0,0013

NS= Not Significant; SD=Standard Deviation; p≤ 0,05

For Total Coliforms in carcasses and knives, significant differences were found, where abattoir 2 got the highest values. While, there were no significant differences for workers' hands and tables variables (Table 3).

Table 3. Total Coliforms Microorganisms for carcasses, knives, workers' hands and tables

Total Coliforms (log_{10} CFU/mL ± SD)	Abattoir 1	Abbatoir 2	Abattoir 3	*P - value*
Carcasses	1,39 ± 0,63[a]	2,1 ± 0.43[c]	1,86 ± 0,62[b]	0,0000
Knives	1,34 ± 0,36[a]	2,0 ± 0,60[c]	1,75 ± 0,60[b]	0,0012
Workers' hands	1,96 ± 0,16	2,1 ± 0,48	2,05 ± 0,36	0,0873NS
Tables	1,95 ± 0,33	1,9 ± 0,44	2,03 ± 0,69	0,0607NS

NS= Not Significant; SD = Standard Deviation; p ≤ 0,05

There were significant differences (p ≤ 0.05) in fecal coliforms analyses for carcasses and knives, especially in abattoir 2 with the highest values exceeding the MLP. In contrast, there were no significant differences on worker's hands and tables (p ≥ 0.05) (Table 4).

Table 4. Fecal Coliforms Microorganisms for carcasses, knives, workers' hands and tables

Fecals Coliforms (log_{10} CFU / mL ± SD)	Abattoir 1	Abattoir 2	Abattoir 3	*P* – value
Carcasses	1,4 ± 0,58[b]	1,4 ± 0,65[c]	1,25 ± 0,77[a]	0,0002
Knives	1,38 ± 0,52[b]	1,6 ± 0,41[bc]	1,87 ± 0,63[a]	0,0239
Workers' hands	1,33 ± 0,69[b]	1,8 ± 0,36[b]	1,42 ± 0,57[a]	0,0052
Tables	1,61 ± 0,44	1,3 ± 0,57	1,9 ± 0,73	0,3923NS

NS= Not Significant; SD = Standard Deviation; p ≤ 0,05

4. Discussions

In this study the initial pH (45 min) average was 6,62; this value is slightly higher to the reported by Maltin *et al.* (2003) who mentioned that in the early *post mortem* period when lactate accumulation does not lead a reduction in pH 6,0 – 6,4. Muscle pH in well-fed animals and pre-slaughter fasting is close to neutrality (6,8 to 7,2) (Forrest, 1975, Lawrie, 1985).

The ultimate pH (24 h) mean for the current research was 5,84, which matches with Komprda *et al.,* (2012) reported that a pH for lambs at 24 h after slaughtered between 5,8 - 5,74, and Kuchtík *et al.,* (2012), between 5,63 – 5,77. The pH found in this study decreased appropriately from 6,27 to 5,70 after slaughtering and, this fall is suitable for acceptable meat characteristics by consumers according to Devine *et al*., 1993. Forrest (1975) and Lawrie (1985) mentioned in their research that ph decreased at 24 h *post mortem* to a rank from 5,4 to 5,5; this values are slightly lower than those reported by the authors mentioned above.

The lowest pH found in this study was 5,70 and the highest was 6,27. Both results are included in the rank of 4,5 to 9,0, where the pH has a significant effect on viability and microorganisms' growth, especially Gram negative bacteria. When it is below it limits not only the cells growth, but also they lose viability (Ray and Bhunia, 2008). This parameter measure is used as an indicator to evaluate its shelf life and quality.

Significant differences were found in color characteristics L*, a* and b* (Table 1). L* values in this research were from 39,34 to 31,61 similar to Mariezcurrena *et al.* (2013), who reported a value of 35,95 for imported meat from New Zealand. Novelo *et al.* (2008) who evaluated the effect of refrigeration temperature and airing period length on Holstein steers' meat quality obtained a value of 36,1 at 24 h after slaughtering. All these investigations suggested that these L* values are appropriated for consumers.

On the other hand, one of the most important qualities in purchase decision is the cherry red color (a*), which is associated with freshness and quality (Brewer *et al.*, 2001, Mancini and Hunt, 2005). In this research significant differences were found in a* among slaughterhouses, however, a* rank values were 11,54 to 14,50, according to Mariezcurrena *et al*. (2013) for national meat 14,37 and lower than those reported by Vignola (2009) 15,41. Positive values of a* indicates red color and negative values of a* indicates green, between them brown color is found. The last two are undesirable colors for consumers. These values are designated by the chemical state and type of myoglobin present on meat. Consequently, it is suggested that color results pointed that myoglobin exists in an appropriate chemical state for a suitable meat color, otherwise, a* values related with brown color are derived from oxidation states of myoglobin, while more myoglobin oxidation is existed more metmyoglobin concentration is found, one of the factors that promoted these values is the presence of oxygen, which promoted the growth of microorganisms. In this work the meat had a proper red color and microorganisms' counts were under Official Standards.

Yellowness (b*) values found in this study were 6,39 to 8,74 similar to those reported by Mariezcurrena *et al.* (2013) for national meat 6,13 and for imported meat 7,41, and with Vignola *et al.* (2009) that recorded a value of 6,95. b* indicates the position between primary colors Y/B (yellow/blue) (Saláková, 2012). Positive values of b* indicates yellow color and negative values of b* indicates blue.

When b* increased L* decreased, which suggests that increases in glycolytic potential promote acidity, paleness (lower L*), and yellowness (greater b*) (Meadus and MacInnis, 2000). Glycolytic potential indicates the capacity for anaerobic metabolism by accounting for the various substrates found in muscle that can be converted to lactic acid (Mancini and Hunt, 2005).

Tenderness values obtained in the current work were from 3,01 kg_f to 4,93 kg_f, these results matched with Novelo *et al.* (2008) who recorded a value of 3,2 kg_f and were lower than those reported by Mariezcurrena *et al.* (2013) for national meat 5,0 kg_f. Boleman *et al.* (1997) classified meat as "very tender" when shear force with a Warner-Bratzler cell is lower than 3,59 kg_f, while when it is higher than 5,90 kg_f is considered as "tough". According to the above mentioned all the studies described above had tender meat.

Tenderness is considered the main meat quality parameter and acceptance. According to MLA (2006) a meat with a mean value of 3,5 kg_f is accepted by consumer. In addition to this, Alliance Group Limited (2010) stated that a meat with a shear force of greater than 11 kg_f is considered tough.

Finally, over time meat is tenderer due to bacterial load. It should be noted that in this work was not evaluated shelf life.

Carcasses and slaughterhouse sanitary characteristics

The results of this study showed that mesophilic aerobic microorganisms loads are suggested as relevant because of it was sampled immediately after slaughtering. Similar reports, as Desdemona *et al.* (2011) quantified over rib and hindquarter with 1,2 and 3,6 $\log_{10}$ CFU / g, respectively, even though it was at 24 h *post mortem* and cooling under sanitary conditions of Type Federal Inspection (TIF). In this report it was obtained 3,59 $\log_{10}$ CFU /mL, with no cooling conditions. Including a higher attachment to European Union Council and Parliament Regulations will improve these bacterial loads as Sumner *et al.* (2002) estimated with 2,8 $\log_{10}$ CFU/mL who attached to this regulation.

Official Mexican Normativity for Total Coliforms LMP it is not considered for fresh meat but to include it could suggest estimation for some Bacterial Generous such as *Citrobacter and Enterobacter.* This kind of contamination could stay in the meat storage at cooling conditions producing some toxins. Then, it is suggested to include it or to establish LMP for these values in row meat. Regarding to Total Coliforms Quantification there is not enough scientific reports useful to compare the present results.

Official Mexican Standard (NOM) don't include Fecal Coliform Testing for raw meat, this is estimated only in cooked meat because of their termotolerancy. Then fecal coliforms couldn't survive to thermal conditions during the cooking. However, this project includes not only Fecal Coliform Testing, but also Total Coliform Testing, which is used as a presumptive testing. In fact, Total Coliform medium used could allow some other no Coliform Genus (i. e. *Aeromonas)* that could produce important toxins. Therefore, it is proposed that the relevant mesophilic species from this Genus must be studied. On the other hand, even though that Fecal

Coliforms were found in living (worker's hands) and not living surfaces (knives and tables) and that these values were close to NF V08-060 Regulations. It didn't ensure fecal contamination because they has been sampled after their use in slaughtering procedures.

However, the results of this study are in the MPL for the Regulations mentioned, therefore, for a second time a tracing of Good Manufacture Practices will improve sanitary characteristics found in carcasses, enamelware and workers' hands.

5. Conclusions

In conclusion, physicochemical characteristics, mainly pH has been necessary for an acceptable meat quality by the consumer. However, microbiological characteristics require a shared responsibility among productive sector and government in order to regulate sheep carcasses production at the Central Highlands of the State of Mexico.

In spite of the overpassed MPL Official Mexican Standards found, the meat physicochemical characteristics haven't been affected. Consequently, It has been recommended an adequate cooking for the consumers and that the suggested shared responsibility includes more Official Standards for more potential meat pathogens identification.

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Corresponding Author:
Dra. Ana Tarin Gutiérrez Ibáñez
Facultad de Ciencias Agrícolas
Universidad Autonoma del Estado de México
E-mail address: atarini@uaemex.mx

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