



Assessment of carcass contamination in a slaughterhouse in the governorate of Blida, Algeria

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ABSTRACT

The initial contamination of meat occurs during slaughter procedures. The objective of this study was to evaluate the hygiene in a slaughter establishment by evaluating the surface contamination of the carcasses and the level of hygiene of the workforce, the environment and the equipment. Altogether, 122 samples were taken, (50 from carcasses and 72 from personnel, equipment and environment). The state of cleanliness of animals was assessed for 125 sheep and 150 cattle. Bacteriological analyses conducted were the enumeration of total coliforms, thermo-tolerant coliforms and *Escherichia coli* and the detection of *Salmonella*.

The carcasses were significantly contaminated with bacterial hygiene indicators and there were no significant differences ($P > 0.05$) in contamination levels between the carcass species, or between the days of the weeks or the microbial groups enumerated. Evaluation of animals' cleanliness showed that 68% of the examined sheep were dirty or very dirty, and 91.33% of the cattle were lightly soiled or dirty. Examination of the contamination of personnel, equipment and the environment between the start and the end of the week did not reveal a significant difference ($P > 0.05$). In order to minimize the contamination of carcasses at the slaughterhouse level, it is recommended to apply good hygiene practices.

1. Introduction

Ensuring food safety at all levels of the food production chain has become a fundamental priority for the food industry. Meat is an excellent source of animal protein, but in addition to the requirements for its nutritional and taste qualities, health quality is essential. Meat is a highly perishable foodstuff the hygienic quality of which depends on the one hand on contamination during slaughter and cutting operations and on the other hand on the development and growth of contaminating microbiota during cooling, storage and distribution ([Dennai *et al.*, 2001; El Hadeef El Okki *et al.*, 2005; Salifou *et al.*, 2013).

The veterinary controls in force at the slaughterhouse level provide some guarantee of the meat's

hygienic status. The controls focus more on animal health compliance that results in healthful meat for consumption, i.e detection of animal diseases that can be transmitted to humans (Sadoud, 2017). Studies have shown that it is the microbial hazards present primarily in healthy animals that are the greatest source of risk to human health, such as *Salmonella enteritidis*, *Campylobacter jejuni*, *Escherichia coli*, *Clostridium perfringens*, *Yersinia enterocolitica* and *Listeria monocytogenes* (FOA, 2006).

In fact, surface contamination of meat mainly takes place at the slaughterhouse despite efforts made by veterinary services to ensure safe meat (Sadoud, 2017). This contamination is, therefore, not desirable, but inevitable. From the point of view

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of legislation, carcasses of slaughter animals are not subject to food safety criteria at slaughterhouse level, i.e., to criteria for which the thresholds must imperatively be respected to place the product in the market. However, the evaluation of surface contamination of carcasses reflects the level of hygiene of the processes and allows corrective actions.

The objective of this study is to assess and determine the surface contamination of the carcasses and the potential sources of contamination in one facility, in order to evaluate the level of hygiene of the slaughterhouse and the cleanliness of live animals.

2. Materials and Methods

2.1 Samples

One hundred and twenty-two samples were taken as follows, 50 from carcasses (25 cattle and 25 sheep); 20 from personnel hands, 20 from personnel clothing (shirts); 16 from knives, 16 from axes and 08 from walls (building).

The visual assessment of the state of cleanliness of animals before slaughter concerned 125 sheep and 150 cattle.

2.3 Sampling

Five carcasses of each species were examined per day each week. Every week, one day of the five days of slaughterhouse exploitation, was dedicated to collecting samples. The choice of carcasses was made randomly.

- Surface swab samples were taken from the surface of freshly slaughtered carcasses declared fit for consumption after health inspection and before the start of drying. The non-destructive method (swabbing) was carried out using abrasive household sponges with a dimension of 5 cm × 5 cm, or 25 cm².
- Two zones were swab sampled per half-carcass (sides and shoulder), i.e., 4 per carcass, making a total area of 400 cm² delimited by a plastic template.
- The sponges from each carcass were placed in the same identified sterile Stomacher bag, supplemented with 100 ml of buffered peptone water. The bag was then hermetically sealed, and placed in a cooler.

Samples from hands, shirts, knives, axes and walls were taken at the beginning and end of the week, using the double swab method. This entailed

a first swab within the template using a cotton tipped swab stick soaked in buffered peptone water followed by a second dry swab within the delimited surfaces. The two swabs were placed aseptically in the same tube with buffered peptone water. The samples were stored in a cooler and sent to the laboratory.

Visual assessment of the state of cleanliness of the animals before slaughter concerned 125 sheep and 150 cattle. Classification of the cattle was based on a grid of four cleanliness classes, A to D (*Bastien et al.*, 2006). The grading of sheep was based on a grid developed on the model of the grid for large cattle but adapted for sheep. It was made up of four classes A to D but took into account dry and wet soiling (*Evrat-Georgel*, 2013). The state of humidity was assessed by palpation of the sheep's fleece.

2.4 Method of microbiological analysis

Upon receipt at the laboratory, each sample was homogenized in a stomacher for ten minutes. The resulting suspension was directly and aseptically poured into an identified sterile vial, this was the stock suspension.

From the stock suspension, a series of decimal dilutions (10^{-1} to 10^{-6}) was carried out in buffered peptone water (IPA®).

2.5 Enumeration of total coliforms and thermotolerant coliforms

The coliform count was carried out by deep-seeding into Petrie dishes containing crystal violet and neutral red agar (VRBL). The dishes were incubated separately, one at 30 °C for 24 hours for the enumeration of total coliforms, and the other at 44 °C for 24 hours for the enumeration of thermotolerant coliforms.

2.6 Enumeration of *Escherichia coli*

The Petrie dishes positive for thermotolerant coliforms at the level of two successive dilutions were retained. A determined number of five characteristic colonies on each of the selected dishes were subcultured, with a view to making a biochemical identification of pure cultures.

2.7 *Salmonella* detection

After preparing the decimal dilutions, the remainder of the stock suspension was incubated at 37 °C for 24 hours for pre-enrichment. Enrichment was performed by adding 1 mL of pre-enrichment in Selenite

Broth and incubating 37 °C for 24 h. Isolation on Hek-toen agar, incubate at 37 °C for 24 h.

After purification of the isolates, we performed biochemical identification with the Api 20^E gallery.

2.8 Statistical analysis

Statistical analyses of data were performed using SPSS version 21 software. Results were subjected to analysis of variance (ANOVA) for multiple comparison tests. The level (of $p < 0.05$) was considered the significant

3. Results

Evaluation of carcass microbiological quality

The results of the enumeration of bacteria indicative of hygiene on bovine and ovine carcasses are shown in Table 1.

The week of sample collection had no significant effect ($P > 0.05$) on the contamination of bovine or ovine carcasses.

For each microbial group enumerated, only non-significant differences ($P > 0.05$) were noted between bovine carcasses and ovine carcasses, except for thermo-tolerant coliforms during the third and fifth week, which did differ significantly (Table 1).

3.1 Assessment of the state of cleanliness of animals

The results of the assessment of the state of cleanliness of sheep and cattle are reported in Table 2.

The majority of sheep (68%) slaughtered were classes C or D (dirty and very dirty, respectively); while about 91% of the cattle examined were classes B or C (lightly soiled and dirty, respectively).

3.2 Evaluation of the hygiene of the workforce, the equipment and the environment

The results of the enumeration of bacteria indicative of personnel hygiene (hands and shirts), equipment surfaces (knives, axes) and the environment (walls) of the slaughterhouse are shown in Table 3.

Table 1. Means ($\pm$ standard deviation) of microbial loads in bovine and ovine carcasses over five weeks

	W1	W2	W3	W4	W5
Total coliforms					
BC	4.713 $\pm$ 1.234 ^{aA}	4.717 $\pm$ 1.139 ^{aA}	5.056 $\pm$ 0.452 ^{aA}	5.448 $\pm$ 0.461 ^{aA}	5.382 $\pm$ 0.824 ^{aA}
OC	5.049 $\pm$ 0.470 ^{aA}	5.353 $\pm$ 0.990 ^{aA}	5.363 $\pm$ 0.600 ^{aA}	4.889 $\pm$ 0.506 ^{aA}	5.718 $\pm$ 0.476 ^{aA}
Thermotolerant coliforms					
BC	4.533 $\pm$ 1.580 ^{aA}	4.584 $\pm$ 1.393 ^{aA}	3.791 $\pm$ 0.567 ^{aA}	4.582 $\pm$ 1.180 ^{aA}	4.234 $\pm$ 0.728 ^{Aa}
OC	4.172 $\pm$ 0.450 ^{aA}	3.941 $\pm$ 0.989 ^{aA}	5.123 $\pm$ 0.802 ^{aB}	4.332 $\pm$ 0.800 ^{aA}	5.289 $\pm$ 0.586 ^{aB}
Escherichia. coli					
BC	3.376 $\pm$ 0.973 ^{aA}	4.038 $\pm$ 0.962 ^{aA}	3.411 $\pm$ 0.790 ^{aA}	4.000 $\pm$ 0.862 ^{aA}	3.577 $\pm$ 0.685 ^{aA}
OC	3.592 $\pm$ 0.690 ^{aA}	3.122 $\pm$ 0.624 ^{aA}	4.126 $\pm$ 0.571 ^{aA}	3.780 $\pm$ 0.512 ^{aA}	4.134 $\pm$ 0.605 ^{aA}

Legend: Values are in log CFU/cm²; OC: ovine carcasses, BC: bovine carcasses, W: weeks.

For each microbial group, values followed by a different lowercase letter within the same row are significantly different ($P < 0.05$) and values followed by a different uppercase letter within a row. The same column is significantly different ($P < 0.05$).

Table 2. Assessment of the state of cleanliness of the animals presented for slaughter

		Class A	Class B	Class C	Class D
Sheep presented for slaughter (n=125)	n	15	25	46	39
	%	12%	20%	36.8%	31.2%
Cattle presented for slaughter (n=150)	n	10	112	26	02
	%	6.66%	74.66%	17.33%	1.33%

Table 3. Means ($\pm$ standard deviation) of the microbial loads of the personal (hands, shirts), equipment (knives, axes) and environment (wall) of the slaughterhouse. Values are in log CFU/cm²

	First day of week (Sunday)	Last day of week (Thursday)
Personnel hands		
Total coliforms	5.098 $\pm$ 0.065 ^a	4.930 $\pm$ 0.565 ^a
Thermotolerant coliforms	4.635 $\pm$ 0.392 ^a	4.381 $\pm$ 0.756 ^a
<i>Escherichia. coli</i>	3.700 $\pm$ 0.398 ^a	3.757 $\pm$ 0.637 ^a
Shirts		
Total coliforms	3.487 $\pm$ 0.353 ^a	3.866 $\pm$ 0.642 ^a
Thermotolerant coliforms	3.011 $\pm$ 0.259 ^a	3.158 $\pm$ 0.414 ^a
<i>Escherichia. coli</i>	2.478 $\pm$ 0.610 ^a	2.647 $\pm$ 0.466 ^a
Knives		
Total coliforms	4.583 $\pm$ 0.434 ^a	4.623 $\pm$ 0.393 ^a
Thermotolerant coliforms	3.792 $\pm$ 0.199 ^a	3.605 $\pm$ 0.316 ^a
<i>Escherichia. coli</i>	2.752 $\pm$ 0.131 ^a	2.783 $\pm$ 0.147 ^a
Axes		
Total coliforms	4.482 $\pm$ 0.417 ^a	4.702 $\pm$ 0.180 ^a
Thermotolerant coliforms	3.559 $\pm$ 0.384 ^a	3.811 $\pm$ 0.258 ^a
<i>Escherichia. coli</i>	2.857 $\pm$ 0.098 ^a	2.684 $\pm$ 0.151 ^a
Walls		
Total coliforms	4.286 $\pm$ 0.151 ^a	2.840 $\pm$ 0.976 ^a
Thermotolerant coliforms	4.119 $\pm$ 0.231 ^a	2.418 $\pm$ 0.570 ^b
<i>Escherichia. coli</i>	2.974 $\pm$ 0.273 ^a	1.342 $\pm$ 0.542 ^b

For each microbial group, values followed by a different lowercase letter within the same row are significantly different ($P < 0.05$).

Between the first and the last working days (respectively Sunday and Thursday) contamination of the personnel (hands of the personnel and shirts), equipment surfaces the material (knives and axes) and the environment (slaughterhouse walls) did not differ significantly ($P > 0.05$), except for walls, for these, significant differences ($P < 0.05$) were recorded for thermo-tolerant coliforms and *Escherichia. coli*.

4. Discussion

Ensuring food safety at all levels of the production chain has become a fundamental priority for the agro-food industries. Currently food hygiene is based on risk analysis. For meat hygiene, slaughter is considered the stage where the greatest opportunities for contamination exist (Hammoudi et al., 2013) and so the slaughterhouse is a strategic point of intervention for the protection of human health. Strict monitoring

of good slaughter hygiene practices is essential in preventing microbial contamination of carcasses. In some countries, slaughter animal carcasses are not subject to criteria for which thresholds must be met, but rather they are subject to process hygiene indicator criteria, the exceeding of which does not require withdrawal measures but corrective actions relating to process hygiene (OJEU, 2005). In Algeria; meat inspection at slaughter establishment level is based on visual examination, palpation and compulsory incision of specified organs in order to exclude from consumption meat that would present a danger to the consumer. However, despite the efforts made by the veterinary services to ensure safe meat, hygienic conditions remain far from optimal and the surface contamination of carcasses is significant (Nouichi and Taha Mossadak, 2009 ; Harhoura et al., 2012; Hammoudi et al., 2013; Benaissa et al., 2014).

In the absence of Algerian legislation for process hygiene criteria, we referred to the European Union standards which recommend the enumeration of *Enterobacteriaceae* with a lower limit m of

1,5 log CFU/cm² and an upper limit M of 2,5 log CFU/cm² (OJEU, 2005). Poor surface quality (in terms of hygiene) of sheep and bovine carcasses has been reported by several studies at the national level (Nouichi and Taha Mossadak, 2009 ; Harhoura *et al.*, 2012; Hammoudi *et al.*, 2013; Bennadji *et al.*, 2013; Benaissa *et al.*, 2014). According to Doulgeraki *et al.* (2012), the bacterial spoilage of meat depends on the initial number of microorganisms, the time / temperature combination of storage conditions and the physico-chemical properties of the meat. Contamination occurs mainly as a result of poor hygienic and handling conditions in slaughterhouses (Schlegelová *et al.*, 2004).

This lack of hygiene was highlighted by the current study that showed there was no significantly measurable difference in hygiene between the days of sampling. Earlier Bennadji *et al* (2013), showed that hygiene was sufficient on Saturday and Sunday, acceptable on Monday and insufficient on the last three days of the week. The sufficiently hygienic situation as we recorded during (Saturday and Sunday) appeared to be the result of the efficient cleaning carried out at the end of the week.

The results also showed there was no significant difference between the contamination of sheep and bovine carcasses. This was probably due to the slaughtering process for sheep and cattle at the slaughter establishment visited where we noted that the slaughter and the start of skinning took place on the floor for both species. Operators manually tear off the skin. This practice forces them to simultaneously touch the fleece and the carcass. This finding is supported by the study by Sadoud (2017) in the Chelf region who reported that slaughter takes place in fixed stations, so the animal is bled, skinned and eviscerated in the same place. In addition, slaughterhouses are, most of the time overcrowded, which promotes contamination. In the study by Bakhtiary *et al* (2016) in Iran where Halal slaughter is carried out reported the bacterial diversity of environmental samples in the sheep slaughter line was higher than that of cattle, probably due to manual slaughter of sheep being practiced on the ground and transmissible contamination via fleece from one animal to another was transmissible. In the cattle slaughter line, all slaughter processes were carried out on a production line with vertical rail dressing and automatic skin removers (Bakhtiary *et al.*, 2016). Contamination of carcasses with *Escherichia coli* can be of concern. Although these bacteria are commensal

to the gastrointestinal tract of many animals, some strains that can be very pathogenic including Shiga-toxin producing *E. coli* STEC. The transmission of these pathogens to humans occurs mainly through the ingestion of food including meat contaminated with digestive contents or bovine feces (Chaucheyras-Durand *et al.*, 2016).

Contamination of carcasses can also be explained by contamination of the animals themselves, i.e. the skin, which is often soiled with various dirt, mud or feces can be a source of contamination. In the present study, cleanliness assessment of sheep showed the majority of the animals were classes C or D (dirty and very dirty, respectively); while the cattle were mostly classes B or C (slightly soiled and dirty, respectively). According to the FAO (2006), sheep fleeces can bring large amounts of dirt and feces into the slaughterhouse. Contamination of sheep carcasses cannot be avoided when the fleece is very dirty. Likewise for bovine carcasses, the skin is a source of contamination. According to Xianqin *et al* (2015) and Dickson and Acuff (2017), minimizing skin contamination or decontaminating the skin could reduce subsequent contamination of the carcass.

Skinning and evisceration are the two most influential steps that can contaminate carcasses and equipment with intestinal bacteria (Lerma *et al.*, 2013).

This study confirms the probable participation of personnel, surfaces and the slaughter environment in the final bacterial load of the carcass. During our presence on the site, we noted during the slaughter some anomalies which can be implicated in general hygiene faults and carcass contamination. We noticed that the staff did not wear appropriate work clothes. The clothes they wore were neither washed nor changed during the entire period of our study. Indeed, hands, hair, beards, and aprons can harbour many microorganisms which can pass very easily to the surface of carcasses by direct contact or by splashes (Labadie, 1999). At the bleeding level, the operator slaughters the first animal, wipes the blade of the knife used on the fleece of the slaughtered animal, and repeats the same gestures to bleed each animal without rinsing his hands or the knife used. In fact, in most of our slaughterhouses, the equipment (knives and axes) is just rinsed at the end of the day (Benaissa *et al.*, 2014). According to Labadie (1999), hooks, storage bins and all equipment (knives, saws, cleavers) that come into contact with meat are soiled by microorganisms. It is essential to remember the fact that each contact brings additional contamination.

The presence of blood, and fat from meat waste on the ground and on the walls contributes to the contamination of carcasses. This state of affairs was reported by Benaissa et al (2014) where poorly designed wall coverings with crevices and cracks that were difficult to clean were nests for microorganisms.

It is very likely that all these unconventional behaviours and the poor hygiene of the environment contributed to the poor hygienic quality found in the carcasses.

5. Conclusion

In order to guarantee meat safety and thus protect consumer health, it is imperative to control the food from barn to table. The slaughterhouse is one of the major critical points in the meat product production chain, and is where biological risks are probably the most worrying. However, the application of good practices and general hygiene can considerably limit microbial contamination of carcasses.

Procena kontaminacije trupova u klanici u pokrajini Blida, Alžir

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INFORMACIJE O RADU

Ključne reči:

Goveda
Životna sredina
Higijena
Klanje
Ovce

APSTRAKT

Inicijalna kontaminacija mesa se dešava tokom postupka klanja. Cilj ovog istraživanja je bio da se proceni higijena u objektu za klanje proverom površinske kontaminacije trupova i nivoa higijene zaposlenih, životne sredine i opreme. Ukupno su uzeta 122 uzorka (50 sa trupova i 72 od osoblja, opreme i životne sredine). Stanje čistoće životinja procenjeno je za 125 ovaca i 150 goveda. Bakteriološke analize su uključivale broj ukupnih koliforma, termotolerantnih koliforma i *Escherichia coli* i otkrivanje salmonela.

Trupovi su bili značajno kontaminirani indikatorima higijene bakterija i nije bilo značajnih razlika ($P > 0,05$) u nivoima kontaminacije između vrsta trupova, niti između dana u sedmici ili popisanih grupa mikroba. Procena čistoće životinja pokazala je da je 68% ispitanih ovaca bilo prljavo ili veoma prljavo, a 91,33% goveda je bilo slabo zaprljano ili prljavo. Ispitivanje kontaminacije osoblja, opreme i životne sredine između početka i kraja nedelje nije otkrilo značajnu razliku ($P > 0,05$). Da bi se kontaminacija trupova na nivou klanice svela na minimum, preporučuje se primena dobre higijenske prakse.

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