

A C OMPARISON BETWEEN SWABBING AND EXCISION SAMPLING METHODS FOR BACTERIAL LOAD OF CARCASSES AT SLAUGHTERHOUSES IN KHARTOUM STATE

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المستخلص

اجرى هذا البحث لتقييم فعالية طريقة التقطيع لجزء من سطح اللحم مقابل طريقة المسحة كطرق لتقييم التلوث البكتيري من ذبائح الأبقار وأيضا من أجل التوصية بمنهجية محددة لإستخدامها في مراقبة وتحسين سلامة ونوعية اللحوم، كالحمل البكتيري للذبائح في مسالخ ولاية الخرطوم.

أخذت عينات المسوحات والقطع من ثلاثة مواقع من ذبائح الأبقار وهي الجناح ، الصدر ، الرقبة بعد مرحلة نزع الأحشاء من الذبائح. بمجموع 240 عينة من 40 ذبيحة من الأبقار (3 عينات مسوحات و 3 عينات قطع من كل ذبيحة) كل العينات تم تجهيزها لحساب العد الحي بإستخدام طريقة مايلز أند ميزرا.

تراوح متوسط العد الحي للبكتريا بين 3.19 إلى 3.5 في السم² وطريقة التقطيع وجدت أنها الأكثر فعالية لعد البكتريا مقارنة بطريقة المسوحات.

متوسط العد الحي للبكتريا لعينات المسوحات من منطقة الجناح ، الصدر والرقبة كان 3.2125، 3.1925 و 3.225 على التوالي، ومتوسط العد الحي للبكتريا لعينات القطع كان 3.48، 3.4525 و 3.505 على التوالي.

وجد فرق معنوي كبير (القيمة الإحتمالية أقل من 0.05) بين طريقتي أخذ العينات بالمسوحات والتقطيع من منطقة الجناح ، الصدر والرقبة، كما وجد أن القيمة الإحتمالية هي 0.003786، 0.005563 و 0.00266 على التوالي.

العد الحي للبكتريا من المسوحات و القطع في المناطق المختلفة كشف عدم وجود إختلاف معنوي بين تلك المواقع (القيمة الإحتمالية أكبر من 0.05)، ونتيجة القيمة الإحتمالية كانت 0.965701 و 0.229126 على التوالي.

أظهرت النتائج بأن طريقة التقطيع تعد البكتيريا أفضل من طريقة المسوحات و وهى بالتالي تعتبر الطريقة الأكثر فعالية لعد البكتيريا من ذبائح الأبقار من المسالخ في ولاية الخرطوم.

Abstract

The objectives of this study is to evaluate the effectiveness of excision of a part of meat surface versus swabbing as methods for the assessment of bacterial load of bovine carcasses and also to recommend a harmonized methodology to be used in monitoring and improving the safety and quality of meat as for as bacterial load of carcasses at slaughterhouses in Khartoum state.

Swabbing and Excision samples were taken from three sites of bovine carcasses which are flank, brisket and neck after the stage of carcasses evisceration.

A total of 240 samples were taken from 40 bovine carcasses (three swabs samples and three excision samples from each carcass), all collected samples were processed for viable counts using miles and misra method.

Mean viable counts ranged from 3.19 to 3.5 cm^{-2} for bovine carcasses and excision method generally recovered bacteria more than swabbing method, the mean viable counts of swabbing from flank, brisket and neck are 3.2125, 3.1925, and 3.225 respectively. And the mean viable counts of excision from these sites are 3.48, 3.4525 and 3.505, respectively.

There is significant difference ($P\text{-value} < 0.05$) between swabbing and excision sampling methods in the flank, brisket and neck, $P\text{-value}$ is 0.003786, 0.005563, and 0.00266, respectively.

Swabbing viable counts at the different sites revealed there is no significant difference ($P\text{-value} > 0.05$) between the different sites and excision viable counts at the different sites also revealed there is no significant difference ($P\text{-value} > 0.05$) between the different sites, $P\text{-value}$ is 0.965701 and 0.229126, respectively.

The results showed that excision method generally recovered bacteria more than swabbing method; therefore, excision method proved to be the most effective technique for bacterial recovery from bovine carcasses at slaughterhouses in Khartoum state.

Key words: Swabbing, Excision, Bovine, Carcasses, Bacterial load, Slaughterhouses, Khartoum State.

Introduction

Traditional visual inspection of carcasses by trained personnel is not an effective strategy for protecting Sudanese consumers against meat-borne infections, physical and chemical hazards because the presence of a meat inspection system examines grossly apparent abnormalities during the ante-mortem and post mortem examination.

Microbial food safety has emerged to be a global concern (Narrung and Buncic, 2008; Sofos, 2008). In response to the increasing number of foodborne illnesses, governments all over the world are intensifying their efforts to improve food safety (Orriss and Whitehead, 2000; Anonymous, 2002; Schlundt, 2002; Codex Alimentarius Commission, 2003; Wallace *et al.*, 2005).

The hazard analysis and critical control point (HACCP) system is the most widely used and internationally accepted food safety management system in the world. The main goal of applying HACCP plans in abattoirs is to ensure that animals are slaughtered and dressed under conditions that mean the meat will carry minimal public health risk. (FAO, 2004).

Various sampling methods have been utilized to determine the number of bacteria on the red meat animal carcasses; the principal sampling methods are swabbing and excision, which have found the widest acceptance and use.

Materials and methods

The study was carried out in the most two important slaughterhouses in Khartoum state which are Elkadaro export slaughterhouse and Ghanawa export slaughterhouse.

Collection of samples:

Twenty visits were made to the slaughterhouses from 11 September to 2 December, 2012. Twelve samples (six swabs samples and six excision samples) were taken in each visit. A total of 240 samples were taken from 40 fresh bovine carcasses (6 samples from each carcass).

Sampling methods:

Swabbing sampling:

Swabbing was performed using wool cotton tipped sticks swabs moistened in normal saline for a minimum of five seconds and the moistened swabs was rubbed vertically, horizontally, and diagonally across the sampling site in area of approximately (25cm²). Samples were subsequently placed into cool boxes containing ice packs and transported to the laboratory of the Department of Food Hygiene and Safety, Faculty of Public and Environmental Health, University of Khartoum, for analysis.

Excision sampling:

The same set of carcasses sampled by swabbing were also sampled using an excision and also were taken from each of the three sample sites by cutting an area of 25 cm² (2 mm depth) using a sterile square templates, blade and forceps. Once excised, samples from each site were placed into a separate sterile container. Samples were placed into a cool box containing ice packs and transported to the Laboratory of the Department of Food Hygiene and Safety, Faculty of Public and Environmental Health, University of Khartoum, for analysis.

Microbiological analyses:

All collected samples were processed for bacterial counts. Miles and Misra method was used for viable count as described by (Wilkie F. Harrigan, 1998).

Samples were suspended in 20 ml of sterile normal saline. Ten fold dilutions were then prepared in tubes containing 9 ml of sterile normal saline.

The colonies were calculated using the following formula:

$$\text{Colony forming units (CFU/cm}^2\text{)} = \frac{\text{Average cfu/plate} \times \text{dilution factor} \times 20}{25}$$

Analysis of the results:

Colony counts were transformed into \log_{10} CFU cm^{-2} and to evaluate the significant differences in Viable count between the two methods, one way analysis of variance (ANOVA) was performed using (SPSS 20.0 software) and significant differences were determined at the 5% level ($P < 0.05$).

Results

Analysis of the data showed that:

There is significant difference between swabbing and excision sampling methods in the flank (**P value < 0.05**).

There is significant difference between swabbing and excision sampling methods in the brisket (**P value < 0.05**).

There is significant difference between swabbing and excision sampling methods in the neck (**P value < 0.05**).

Swabbing viable counts at the different sites revealed there was no significant difference between different sites. (**P value > 0.05**).

Excision viable counts at the different sites revealed there was no significant difference between different sites. (**P value>0.05**).

Statistical analysis:

Table 1. Summary of Statistical analysis of swabbing and excision data from the flank site

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
<i>Swabbing</i>	40	128.5	3.2125	0.306763
<i>Excision</i>	40	139.2	3.48	0.014462

Table 2. ANOVA of swabbing and excision data from the flank site

<i>Source of Variation</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
<i>Between Groups</i>	1.431125	1	1.431125	8.910439	0.003786	3.963472
<i>Within groups</i>	12.52775	78	0.160612			
<i>Total</i>	13.95888	79				

*There is significant difference between swabbing and excision sampling methods in the flank (**P-value< 0.05**).

Table 3. Summary of Statistical analysis of swabbing and excision data from the brisket site

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
<i>Swabbing</i>	40	1127.7	33.1925	0.0314558
<i>Excision</i>	40	1138.1	3.4525	0.0017942

Table 4. ANOVA of swabbing and excision data from the brisket site

<i>Source of Variation</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
<i>Between groups</i>	1.352	1	1.352	8.132331	0.005563	3.963472
<i>within groups</i>	12.9675	78	0.16625			
<i>Total</i>	14.3195	79				

*There is significant difference between swabbing and excision sampling methods in the brisket (**P-value < 0.05**).

Table 5. Summary of statistical analysis of swabbing and excision data from the neck site

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
<i>Swabbing</i>	40	129	3.225	0.302436
<i>Excision</i>	40	140.2	3.505	0.023051

Table 6. ANOVA of swabbing and excision data from the neck site

<i>Source of Variation</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
<i>Between groups</i>	1.568	1	1.568	9.634788	0.00266	3.963472
<i>Within Groups</i>	12.694	78	0.162744			
<i>Total</i>	14.262	79				

*There is significant difference between swabbing and excision sampling methods in the neck (**P-value < 0.05**)

Table 7. Summary of Statistical Analysis of swabbing data from the three sites:

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
<i>Flank</i>	40	128.5	3.2125	0.306763
<i>Brisket</i>	40	127.7	3.1925	0.314558
<i>Neck</i>	40	129	3.225	0.302436

Table 8. ANOVA of swabbing data from the three sites:

<i>Source of Variation</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
<i>Between Groups</i>	0.0215	2	0.01075	0.034912	0.965701	3.073763
<i>Within groups</i>	36.0265	117	0.307919			
<i>Total</i>	36.048	119				

*There is no significant difference between the three sites (**P-value>0.05**)

Table 9. Summary of Statistical Analysis of excision data from the three sites:

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
<i>Flank</i>	40	139.2	3.48	0.014462
<i>Brisket</i>	40	138.1	3.4525	0.017942
<i>Neck</i>	40	140.2	3.505	0.023051

Table 10. ANOVA of excision data from the three sites:

<i>Source of variation</i>	<i>SS</i>	<i>DF</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
<i>Between groups</i>	0.055167	2	0.027583	1.492197	0.229126	3.073763
<i>Within groups</i>	2.16275	117	0.018485			
<i>Total</i>	2.217917	119				

*There is no significant difference between the three sites (**P-value>0.05**).

Discussion

This study showed that the excision recovered bacteria more than wool cotton tipped sticks swabs. Our results substantiated these of:

- Morgan *et al.*, (1985) who concluded that excision sampling gave generally higher counts.
- Flisset *al.*, (1991) who concluded that the Stomaching excised skin recovered the highest number of bacteria.

- Ware *et al.*, (1995) who concluded that the results indicated that as the carcass tissue was stored, recovery of bacteria by Swabbing was less efficient than was recovered by excision. Dorsa *et al.*, (1996) found significantly lower bacterial recovery from cotton tipped swabs than from cellulose acetate sponge swabbing, cheese cloth swabbing or excision.
- Gill, C.O. (2001) who also concluded that numbers recovered by swabbing with cotton wool were at the lower end of or below the range of the numbers recovered by the other methods.
- Anonymous (2001) concluded that excision is the most effective technique for bacterial recovery from beef and pork carcasses.
- Hutchison *et al.*, (2005) concluded that poorly correlated linear relationships between swab and excision derived bacterial numbers from near-adjacent carcasses were observed for all three animal species. Who found that the total counts for all species sampled by excision was significantly greater than measured for swabbing.
- Y Ghafiret *et al.*, (2008) concluded that Recovery was significantly lower for the swabbing method in comparison with the destructive method. Bacterial recovery from carcass surfaces may also depend on the animal species (Gill *et al.*, 2001). For example, Gill and Jones (2000) found that bacterial recovery using excision and cellulose acetate sponge swabbing were not significantly different on beef carcasses, however the cellulose acetate sponge recovered significantly less bacteria on pork and lamb carcasses.
- Gill *et al.*, (2001) noted that relationship between the numbers recovered by excision or any selected swabbing technique may differ for different types of noncomminuted.
- Richard *et al.*, (2005) found that several factors that potentially contributed to relatively low and highly variable bacterial recoveries obtained by swabbing were investigated in separate experiments.
- Pearce *et al.*, (2005) found that sampling using the polyurethane sponge represents an equivalent alternative method to excision for the bacteriological sampling of carcass surfaces.
- M. Lindblad (2007) Swabbing with gauze generally recovered bacterial numbers that were comparable with those obtained by excision.

However, to date, a quantitative conversion factor between excision and swabbing has not yet been established. This may be due to the many sources of variation in swabbing data including operator related differences (Snijders *et al.*, 1984), animal species and carcass surface variation (Gillet *et al.*, 2001), the use of different sponge materials (Dorsa *et al.*, 1996; Gill and Jones, 2000; Gill *et al.*, 2001), the presence of inhibitory materials in the sponge (Daley *et al.*, 1995) as well as the time and storage of carcasses before sampling (Lazarus *et al.*, 1977; Ware *et al.*, 1999). With these factors in mind, (Gill *et al.*, 2001) suggest that any assumed relationship between swabbing and excision would be tentative.

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