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# CAMPYLOBACTER FETUS SUBSP JEJUNI IN INTACT SHEEP AND BUFFALO CARCASES (With 2 Tables)

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الكامبيلوباكتر جيوجيناى في ذبائح الأغنام والجامسسوس

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أجربت هذه الدراسة على عدد صدون ذبيحة من الأغنام والجاموس جمعت من مجزر أسيوط بواقع ثلاثين ذبيحة من كل نوع لمعرفة مدى تواجد ميكروب الكامبيلوباكتر جيوجيناى فى هذه اللبائح ولقد تم جمع مائتان وأربعون عينة من كل من الصغرا، الكبد ، لحوم ( الجزء العضلى من الحجاب الحاجز ) والعقد الليمفاوية للامعا، لدراسة توزيع ومدى إنتشار هذا الميكروب فى هذه الأماكن ، وقد ثبت من الفحص الميكروبيولوجي لللبائح تواجد ميكروب الكامبيلوباكتر جيوجيناى بنسبة ١٦ر١١٪ فى كل من ذبائح الأغنام والجاموس التى تم فحصها ولقد تم عزل هذا الميكروب بنسبة ٢٢ر٢١٪ من ذبائح الأغنام أما فى ذبائح الجاموس فكانت نسبة تواجد الميكروب ، ١٠ وقد وجد أن الكامبيلوباكتر يتواجد فى الصغراء ، الكبد والعقد الليمفاويت الميكروب فى نفس الأماكن للبائح الجاموس فكانت ١٢ر٦٪ بالترتيب أما نسبة توزيد عصلا الميكروب فى نفس الأماكن للبائح الجاموس فكانت ١٢ر٢٪ ، ٢٣ر٢٪ ، ٢٣ر٢٪ على التوالدي ولم يستدل على وجود الميكروب فى لحوم اللبائح التي تم فحصها ( الجزء العضلي للحجاب الحاجز ومما هو جدير بالذكر أن الميكروب وجد فى كل من الكبد والصفراء فى أربع من اللبائد عداول مثل هذه اللحوم .

#### SUMMARY

60 freshly slaughtered sheep and buffalo carcases (30 of each) were examined for the presence of Campylobacter jejuni by enrichment and direct plating procedures. Intact gall bladder, liver, muscle (diaphragm) and mesenteric lymph nodes samples were obtained from each animal. The recovery rate of C.jejuni from sheep carcases (23.33%) was higher than that in buffalo carcases (10%). the incidence of the isolated organisms from gall bladder, liver and mesenteric lymph nodes samples of sheep were 16.67%, 10% and 6.67% while the corresponding values in buffalo were 6.67%, 3.33% and 3.33% respectively. C.jejuni failed to be recovered from the examined muscle samples of both animals. The 4 positive livers for Campylobacter were from animals with postive gall bldders. The significance of Campylobacter as a foodborne pathogen was discussed.

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### INTRODUCTION

It is now well established that Campylobacter fetus subsp. Jejuni is a common cause of human enteric disease. Red meats are recognized as a vehicle of salmonellosis but to date, there is little evidence to implicate a widespread association between red meats and Campylobacter infection (BUTZLER and SKIRROW, 1979; SKIRROW, 1982).

In recent year, reports from around the world have demonstrated beyond doubt the importance of C.jejuni and C.coli in human enteritis. There has also been an increasing concern over the role of food animals as reservoirs of these organisms with the implication that Campylobacter diarrhea is a zoonotic infection. However, the organism has been isolated from most common domestic animal species, so it has been inferred that direct transmission of the disease to human might occur via consumption of animal products (BUTZLER and SKIRROW, 1979; GARCIA, et al. 1985). Furthermore, a number of studies have shown that C.jejuni and C.coli are commonly found in healthy as well as diarrheatic animals and that the organisms can be easily isolated from gall bladders and intestinal contents of pigs, sheep and cattle (SMIBERT, 1965; BRYNER, et al. 1972; BLASER, et al. 1980; PRESCOTT and BRUIN-MOSCH, 1981; ROSEF, 1981; MUNORSE, et al. 1983 and GARCIA, et al. 1985).

However, carcases of most other animal providing meat can be contaminated by C.fetus subsp. jejuni. The incidence varies with animal species, but generally, the degree of contamination of carcases seems to be low (STERN, 1981; SVEDHEM and KALJSER, 1981; GILL and HARRIS, 1982b). In this respect, the recovery rate of Campylobacter fetus subsp. jejuni on unwashed 58 pork, 59 lamb and 58 beef carcases was studied by STERN (1981) who recorded that the potential pathogen was present on 38%, 24% and 2% of the examined swine, sheep and beef carcases.

BOLTON, et al. (1982) examined carcases in abattoirs and in butcher's shops. Campy-lobacters were isolated from 32% of beef, 70% of sheep and 56% of pig carcases when sampled at abattoirs but not from any carcases examined in butcher's shops. On the other hand, the level of contaminatin with C.jejuni described by GILL and HARRIS (1982 a) was about one to 10/cm² where the organism was recovered from the flank but not the rump areas. Chilled carcases and deboned veal appeared to be les frequently contaminated with C.jejuni. Besides, small numbers of C.jejuni could be recovered from equipment during the processing of unweaned calves but not after routine cleaning.

TURNBULL and ROSE (1982) revealed the presence of C.jejuni in 1% of 6000 red meat samples examined at retail distribution thus the contamination rate of raw red meat by the organism is in general very low. On the other hand, BRACEWELL, et al. (1985) found that 12.5% of 112 freshly slaughtered pork carcases obtained from three packing plants contained Campylobacter coli while no isolates were obtained from chilled carcases.

In a survey conducted by STERN, et al. (1985) for the presence of C.jejuni and C.coli in 1800 red meat products, the recovery rate was about 5.1% Pork samples yielded Assiut Vet.Med.J. Vol. 22, No. 43, 1989.

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C.coli and other meats yielded C.jejuni. These results provide a baseline for the prevalence of Campylobacter in the selected foods and also support epidemiological data associating mishandled foods of animal origin as a potential vehicle in human gastroenteritis.

GARCIA, et al. (1985) reported the isolation of C.jejuni and C.coli from a total of 525 specimens from 100 slaughtered beef cattle where the organisms were isolated from steers, bulls, heifers and cows. Significantly higher isolation rates were obtained from the gall bladders, large intestines and small intestines than from the livers or lymph nodes.

In Egypt, one article by KHALAF ALLA (1985) revealed the recovery rate of C.jejuni and C.intestinals from cattle, buffalo, sheep and camel carcases. The incidence of C.jejuni in cattle and sheep was the same (12%), while in buffalo and camel it was 8% and 4% respectively. Furthermore, C.fetus subsp intestinals was isolated from buffalo and sheep carcases only.

As C-jejuni is a commensal in the intestines of domestic animals such as cows, swine, sheep and poultry the meat of these animals may become contaminated with the organisms during slaughtering (GRANT, et al. 1980; STERN, 1981 and BLASER, 1982).

This investigation was carried out to study the distribution of C.jejuni among various sites of the digestive tract of sheep and buffalo and to estimate the recovery rate of the potential pathogen on meats.

# MATERIAL and METHODS

60 sheep and buffalo carcases (30 carcases of each) were selected at random from Assiut slaughter house and sampled for isolation of C.jejuni. A total of 240 specimens were collected under sterile conditions in sterile separate plastic bags and transferred directly to the laboratory for processing. The specimens obtained from each animal included intact gall bladder, muscle sample (diaphragm), liver and mesenteric lymph nodes.

By means of hot spatula the surfaces of muscles, livers and defated lymph nodes were sterilized, then by the aid of a forceps and scalpel pieces from deep tissues were taken under sterile conditions and streaked onto Muller-Hinton agar supplemented with 10% sheep blood, trimethoprium 5 mg/liter, vancomycin 10 mg/liter, polymxin B 2500 lU/liter and 0.05 sodium pyruvate (GARCIA, et al. 1985).

Isolation of Campylobacter from the gall bladder was done by inserting sterile pipette through an incision into the gall-bladder after sterilization of its surface, then few milliliters were drawn from the bile and spread over the surface of Muller-Hinton blood agar using surface plating technique.

All inoculated plates were incubated at  $43^{\circ}$ C for 48 h in microaerobic gas mixture of 10% Co<sub>2</sub>, 5% O<sub>2</sub> and 85% N<sub>2</sub> using Campylobacter gas generating kits (Oxoid).

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For the enrichment technique, the prepared organs and bile samples were inoculated into flasks containing the enrichment broth described by ROSEF (1981) which incubated at 43°C in the microaerobic gas mixture. After 24 and 48 h, each inoculated enrichment broth was streaked onto plates of Muller-Hinton blood agar with supplements which incubated at 43°C for 48 h under the same microaerophilic condition. Non swarming suspect colonies resembling those of Campylobacter were subjected to biochemical characteristics according the procedures described by PARK, et al. (1984).

#### RESULTS

Results indicating that the recovery of C.jejuni from slaughtered sheep and buffalo carcases are given in Tables 1 & 2. Of the carcases sampled immediately after slaughter 10 (16.67%) were found to contain C.jejuni. Sheep was found to harbour C. jejuni (23.33%) more than buffalo (10%). The recovery rate of the organism from gall bladder, liver and mesenteric lymph node samples of sheep was 16.67%, 10% and 6.67%, while that of buffalow was 6.67%, 3.33% and 3.33% respectively as presented in Table (1).

C.jejuni failed to be detected in the examined muscle samples of both sheep and buffalo as recorded in Table (1). On the other hand Table (2) revealed that the organism was recovered from both gall bladder and liver samples of 3 sheep and one buffalo.

## DISCUSSION

Compylobacter jejuni is now established enteropathogen that has been responsible for several foodassociated disease outbreaks. The organism is frequently present in the intestinal tract of domestic animals and has been isolated from carcases of slaughtered beef, sheep and swine (ROSEF, 1981; STERN, 1981; HUDSON and ROBERTS, 1982).

The present study demonstrates that 10 out of 60 slaughtered sheep and buffalo (16.67%) are reservoirs of C.jejuni and constitutes potential sources of enteric infection. The incidence of C.jejuni in buffalo is 10% and this figure is much lower than in previous reports on C.jejuni isolation rates of 32% and 50% from beef carcases as reported by BOLTON, et al. (1982) and GARCIA, et al. (1985).

Furthermore the 23.33% incidence in sheep is also lower than that demonstrated by BOLTON, et al. (1982) who recorded an incidence of 56% and the achieved results run parallel with that of STERN (1981) who reported an incidence of 24%. On the other hand, KALAF ALLA (1985) revealed that the recovery rate of C.jejuni from buffalo and sheep was 8% and 12% which appeared to be lower than the reported results.

The isolation of C.jejuni from 6.67% of the examined livers is significant considering the increasing emphasis on beef and sheep livers consumption in human nutrition. However, experimental evidence conducted by SOONATTRAKUL, et al. (1971) has demonstrated that beef and lamb livers are a principal site of Campylobacter infection, and the organism was isolated from the blood of an individual who had eaten such a product.

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The recovery rate of C.jejuni from the gall bladders of 11.67% of the examined animals indicating that the organism is a potential inhabitant of the gall bladders as observed by BRYNER, et al. (1972) and GARCIA, et al. (1985). The high rate of Campylobacter isolation from the gall bladders and liver may partly attributed to the presence of bile which improves the growth of C.jejuni (OOSTEROM, et al. 1981). However, it is likely that there was internal migration of C.jejuni between the gall bladder, the bile duct and the liver either in vivo or soon after slaughter (GARCIA, et al. 1985). In this respect, it is of interest to note that all the 4 positive livers for C.jejuni were from animals with positive gall bladders.

Campylobacter jejuni failed to be detected in the examined muscle samples. Some studies have shown that the incidence of C.jejuni in raw red meat is very low (SEVEDHEM and KAIJSER, 1981; STERN, 1981; TURNBULL and ROSE, 1982).

The results of this study indicate that the contamination level of buffalo and lamb carcases in Upper Egypt were much lower than those reported by researchers in other locations.

Due to the relatively hot weather in Upper Egypt which favour the growth of Campylobacter as reported by SMIBERT (1974) who pointed out that C.jejuni is a micro-aerophilic and requires a minimum temperature of about 30°C thus the presence of the organism in the examined carcases constitutes a public health hazard.

Since Campylobacter fetus subsp. jejuni has been implicated as an agent of foodborne disease, it is important that individuals involved with production, processing or preparation of food be aware of foodborne disease potential of this organism (DOYLE, 1981).

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Table (1)
Frequency distribution of C.jejuni in slaughtered sheep and buffalo.

Animal species	No. of positive animals or specimens/No. tested (%)				
	Animals	Gall bladder	Liver	Muscle	Mesenteric lymph nodes
Sheep	7/30(23,33)	5/30(16.67)	3/30(10)	0/30(0)	2/30(6.67)
Buffalo	3/30(10)	2/30( 6.67)	1/30(3.33)	0/30(0)	1/30(3.33)
Total	10/60(16.67)	7/60(11.67)	4/60(6.67)	0/60(0)	3/60(5)
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Table (2)

Frequency and distribution of C.jejuni in slaughtered sheep and buffalo.

Isolation from single or multiple specimens per animal	No. and % of po	ositive animals Buffalo		
One specimens				
Gall bladder	2	1		
Mesentieric L. node	2	1		
Total	4 (57.14)	2 (66.67)		
Two specimens				
Gall bladder + liver	3 (42.85)	1 (33.33)		