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## STUDIES ON YERSINIA ENTEROCOLITICA MICRO-ORGANISM IN SOME MEAT PRODUCTS

(With 3 Tables)

By

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### دراسات عن ميكروب اليرسينيا إنتيروكوليتكا في بعض منتجات اللحوم

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أجريت هذه الدراسة على عدد ٨٢ عينة من منتجات اللحوم شملت ٥٢ لحم مفروم ، ١٥ لانشون ، ١٥ بسطمره جمعت من السوبر ماركت المختلفه بمدينة أسيوط لمعرفة مدى تواجد ميكروب اليرسينيا إنتيروكوليتكا فيها وطرق العزل البكتيرييه المختلفه . وقد تم عزل الميكروب من تسعة عينات لحم مفروم بنسبة ٣٦ ر ١٧ % ، وعينه واحده من اللانشون بنسبة ٦ ر ٦ % ولم يتم عزله من البسطمره وقد تم تصنيف الميكروب بيوكيميائيا بنوع ٤ وقد تمت مناقشة النتائج وأهمية وخطورة تواجد هذا الميكروب على الصحة العامه وما يجب اتخاذه لمنه انتشار هذا الميكروب .

## SUMMARY

A total of 82 random samples (52 minced meat, 15 luncheon and 15 basterma) from different supermarkets in Assiut City were examined for the occurrence of *Yersinia enterocolitica* by different methods. The organism was isolated from 9 samples of minced meat (17.31%) and one sample of luncheon (6.6%). All isolates belonged to biotype 4. The importance of *Y. enterocolitica* as a public health hazard was discussed.

**Keywords:** Studies, *Yersinia Enterocolitica*, meat products.

## INTRODUCTION

*Yersinia enterocolitica* infection of the gastrointestinal tract have increasingly been reported in human during the past decade (BOTTONNE 1977, KHOL et al., 1977, BLAK et al., 1978; CAPRIOLI et al., 1978). Acute gastroenteritis is the most frequent clinical form of this infection followed by an acute iliac fossa syndrome (pseudoappendicitis), mesenteric lymphadenitis or terminal ileitis). Other clinical conditions described are septicemia, polyarthritits, erythema nodosum and abscesses (TOMA AND LUCETTE 1974). The organism is usually isolated from faeces and less frequently from appendixmesentric lymph nodes, abscess blood, urine and from asymptomatic carrier (SCHIEMANN and TOMA 1978).

Epidemiological study of human *Yersinia* infection has implicated water, animal, food and other environmental sources as reservoir of the organism (MORRIS and FEELEY 1976, LEE 1977, CHRISTENSEN 1982, DELMAS et al., 1982, MEADOWS and SNUDDEN 1982).

Reports from various countries indicate that *Y. enterocolitica* is commonly found in various animal species (LANGFORD 1972, INOUE and KUROSE 1975, TOMA and DEIDRICK 1975, LEE 1977, HUGHES 1979, PEDERSON and WINBLAD 1979).

The aim of our study was to determine the incidence of *Y. enterocolitica* in some meat products (minced meat, luncheon, basterma) by comparative study on different media for isolation and the role of the organism as a public health hazard.

## MATERIAL AND METHODS

## Collection and Preparation of Samples:

Samples of meat products included (52 minced meat, 15 luncheon and 15 basterma) were collected from different supermarkets in Assiut city in sterile plastic bags. The samples were transferred in freeze box to the laboratory with a minimum of delay.

Isolation of *Y. enterocolitica*:

A-11 gm of each sample was placed in a sterile flask, containing 99 ml of selenite broth then incubated at 37°C for 18 hours. From the selenite broth, a loopfull was in-oculated into S.S agar and cefsulodin-Irgasan-Novobiocin (CI N) agar which incubated at 37°C for 24-48 hours. The colony having the morphological character of *Y. enterocolitica* was picked up and streaked into slope agar to be subjected for further identification.

B-One gram of each sample was thoroughly mixed and transferred to 10 ml phosphate buffer saline (PBS, M/15, PH.7.6) which incubated at 4°C for 14 days for enrichment. Enrichment samples were inoculated into cefsulodin-Irgasan-Novobiocin (CIN) agar (oxid) (SCHIEMANN 1979 a, 1979b) and S.S. agar and incubated at 37°C for 24-48 hours.

The criteria used for presumptive identification of *Y. enterocolitica* was a colony having a deep red center "bulleye" surrounded by a transparent border. The edge of the colony was entire or irregular. All presumptive colonies were transferred to nutrient agar slant and stored after incubation at 32°C for 24 hours in a refrigerator at 4°C for later biochemical reactions as shown in table 1. (SCHIEMANN and DEVENISH, 1982), which revealed that the reaction on kligler iron agar, together with tests for urea hydrolysis, and sucrose and salicin fermentation are sufficient for differentiating *Y. enterocolitica* from other organisms that are capable of growing on CIN medium and also for distinguishing pathogenic forms.

The strains were biotyped according to WAUTERS (1970), Table 2.

## RESULTS

The obtained results are summarized in Table (3).

## DISCUSSION

Human infection due to *Y. enterocolitica* have now been reported in numerous countries all over the world. There has been a steady increase in the number of countries which report this infection. In spite of these facts, knowledge of the geographical distribution of this organism is still fragmentary.

The results of this study showed that the incidence of *Y. enterocolitica* among the examined samples of meat products (Minced meat, Luncheon, Basterma) were 17.31%, 6.6% and 0.0% respectively. This rate is nearly similar to those recorded by HANNA et al. (1976) which isolated *Y. enterocolitica* from vacuum packaged beef and lamb also to those studies of SCHIEMANN (1980) who isolated *Y. enterocolitica* from a sample of ground pork 10.8%). ANDERSON et al., (1991) detected *Y. enterocolitica* in 3 out of 24 samples of minced meat (12.5%) on the other hand. ABDEL ALL (1993) isolated *Y. enterocolitica* from minced meat, luncheon, basterma in a rate of 10.34%, 0.0%, 0.0% respectively. The failure of isolation of *Y. enterocolitica* from luncheon and basterma may be due to the fact that curing process play a great effect on the survival and multiplication of the microorganism. The presence of nitrates has also a bacteriostatic effect on the microorganism (CASTELLANI et al., 1955 and Libby 1975). Furthermore, the organism failed to be detected in the examined luncheon and basterma as the result of garlic content on its surface which as a bacteriocidal or bacteriostatic effect on the microorganism (VORLLRATH et al., 1977).

From table 3, it was shown that the incidence of isolation of *Y. enterocolitica* after culturing in phosphate buffer saline then inoculation on CIN agar plates is more than that culturing in selenite broth and inoculation on S.S. agar. These results is similar to those reported by MARTIN and WASHINGTON (1980), SACK et al. (1980) they noticed that none of the usual enteric plating media used for enterobacteriaceae is highly selective or differential for *Y. enterocolitica* such as S.S. agar which will not grow all strains. SCHIEMANN (1979) concluded that CIN agar media was more selective than S.S. agar.

The difference in these results may be attributed to the incorrect hygienic measures during preparing and processing of these products, also to various temperatures used in storing the products as freezing to  $-18^{\circ}\text{C}$  and  $-75^{\circ}\text{C}$  resulted in 7% and 42% cell inactivation respectively (GREEZ and EL-ZAWAHRY, 1984).

## YERSINIA ENTEROCOLITICA & MEAT PRODUCTS

As the refrigeration is commonly used in food stores and private home to prolong the time of storage of perishable food, this treatment gives a reasonable protection against the multiplication of a number of pathogens. *Y. enterocolitica* however, is one of the few human pathogen which is able to grow at refrigerator temperature (NILEHN, 1969 and WAUTERS, 1970); (ZEN-YOJI et al., 1974). So refrigeration favour the growth of *Y. enterocolitica* analagous to the cold enrichment procedure used in isolation of microbial group.

BOYCE et al. (1979) showed that *Y. enterocolitica* enterotoxin is an acid stable and is not inactivated by exposure to 120C<sub>1/2</sub> for 30 minutes or by storage at 4C<sub>1/2</sub> for at least 5 months. They suggested that this toxin is able to with stand gastric acidity and temperature used in food processing and storage. Thus, performed enterotoxin may be capable of causing food-borne intoxication *Health and Welfare Canada 1976 and BLACK et al.*, 1978). So the contamination of meat products with *Y. enterocolitica* during prepaing and processing suggests that attention to hygienic handling practices may be an important preventive measures. Likewise using freezing (not chilling) as well as proper heat treatment would eliminate the risk of infection by this organism.

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Table (1): Typical reactions of pathogenic *Yersinia enterocolitica*

Test	Incubation Temperature	Test Result
Kligler Iron Agar	35C°	Alkaline/acid but no gas or H <sub>2</sub> S positive
Urea hydrolysis	35C°	Positive
Sucrose fermentation	22C°	Positive
Salicin Fermentation	35C°	Negative

Table (2): Wauters biotyping of *Yersinia enterocolitica*

	1	2	3	4	5
Lecithinase	+	-	-	-	-
Indole-	+	+	-	-	-
Trehalose	+	+	+	+	-

Table (3): Incidence of *Yersinia enterocolitica* in some meat products.

Type of Sample	No. of examind Sample	Number of Positive								Biotyping
		Selenite Broth				Phosphate Buffer Saline				
		S.S. Agar		CIN Agar		S.S. Agar		CIN Agar		
		No.	%	No.	%	No.	%	No.	%	
Minced meat	52	6	11.54	3	5.8	2	3.8	9	17.31	4
Luncheon	15	0	0	0	0	0	0	1	6.6	4
Basterma	15	0	0	0	0	0	0	0	0	-