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INCIDENCE OF AEROMONAS HYDROPHILA IN SOME SELECTED FROZEN MEAT PRODUCTS IN ASSIUT CITY

(With Two Tables)

By

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دراسة مدي تواجد ميكروب الايروموناس
هايدروفيليا في بعض منتجات اللحوم المجمدة في مدينة أسيوط

شوكت فتحي ، صباح مصطفي

تضمنت الدراسة فحص ٤٥ عينة من البرجر ، اللحم المفروم والسجق المجمد بواقع خمسة عشر عينة من كل منهم بهدف التعرف علي تواجد ميكروب الايروموناس هايدروفيليا بواسطة المنابت البكتيرية · وبينت النتائج عن تواجد ميكروب الايروموناس هايدروفيليا في جميع العينات بمتوسطات ٣٠ × ١٠٪ ، ٢١ × ١٠٪ ، ٢٩ × ١٠٪ في العينات السابق ذكرها علي التوالي · وكما تم تصنيف ميكروب الايروموناس هايدروفيليا الي ايروموناس هايدروفيليا ايروموناس سوبريا وايروموناس سانيا بنسب مئوية ٣٣ر٣٣٪ ، ٤٠٪ و ٢٦ر٦٧٪ ، ١١ر٤٦٪ ، ٢٦ر٦٧٪ و ٢٦ر٦٧٪ و ١١ر٦٧ و ٦٦ر١١٪ علي التوالي · كما بين البحث الأهمية الصحية لميكروب الايروموناس ·

SUMMARY

45 frozen burger, minced meat and sausage samples (15 each) were examined for the presence of Aeromonas hydrophila by using enrichment and plating procedures. Aeromonas hydrophila was found in all of the examined samples with average count $30x10^7$, $21x10^7$ and $29x10^7$ in the examined burger, minced meat and sausage samples respectivelt. A.hydrophila species was/were identified into A.hydrophila, A.sobria and A.caviae group, where the percentage of their presence in examined samples was 33.33, 40 and 26.67, 46.66, 26.67 and 26.67 and · 26.67, 66.67 and 6.66 respectively.

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INTRODUCTION

Aeromonas hydrophila was recognized as an opportunistic pathogen of low virulence, but recently, it is accepted as a primacy pathogen (LITTEL, et al. 1986). Although the potential of the organism to induce outbreaks of such magnitude is unknown, gastroenteritis caused by A.hydrophila is being recognized more frequently. It has been recognized as a spoiler (LERKE, et al. 1965 and ALUR, et al. 1971) as well as enteric pathogen (BUCHANAN and PALUMBO, 1985).

The organism is capable to grow at refrigerated temperature and has been observed as a part of microflora of fish, milk, poultry, and meat (PALUMBO, et al. 1985 a,b). Increasing interest concerning the possible role of species of the A.hydrophila group (A.hydrophila, A.sobria, and A.caviae) as a cause of human gastroenteritis; both clinical and laboratory investigations have suggested that the species is a significant enteric pathogen (HAZEN, et al. 1978; GRACEY, et al. 1982 and BURKE, et al. 1983). The spoilage potential and pathogenicity of the organism have been correlated to its ability to secrete several extracellular virulence factors including enterotoxins, cytotoxins, haemolysis, lipase and proteases (TRUST and CHIPMAN, 1979 and LJUNGH & WADSTROM, 1983).

ENFORS, et al. (1979) recovered A.hydrophila at $10^6/cm^2$ from pure nitrogen-packaged pork cuts but failed to recover the organism from packages with a pure carbon dioxide atmosphere, while PALUMBO, et al. (1985 a) found the organism, often in high numbers, on almost all products of animal origin at the retail level. LIKEWISE, OKREND, et al. (1987) isolated A.hydrophila from all poultry and beef samples and seven of ten pork samples tested.

STERN, et al. (1987) found that the incidence of Aeromonas detected from feces of beef, pig, sheep and turkey was one of 32, none of 22, none of 24 and three of 21 respectively, while TERNSTROM and MOLIN (1987) reported that the percentage of A.hydrophila isolated from raw pork, beef and chicken was 33, 27 and 24 out of 45 samples examined of each respectively.

In the present study a brief survey of some selected retail frozen meat products was conducted for the presence of A.hydrophila.

MATERIAL and METHODS

45 frozen meat products samples were collected from different local retail markets in regular consummer packages in Assiut City, 15 samples from each burger, minced meat and sausage. The samples were analyzed by using enrichment method, where

25 g sample were aseptically transferred to 255 ml of Trypticase soy broth containing 10ug ampicillin/ml and blended for 2 min., then incubated at 28°C for 6 and/or 24 h. After incubation the enrichment culturs were serially diluted up to 10^{-6} in Butterfield's phosphate diluent and spreaded on MacConkey manitol ampicillin agar with a bent glass rod and incubated at 28°C for 18-24 h. The numbers of isolated Aeromonas were estimated and the typical red colonies were picked to triple sugar iron agar and nutrient agar slants. After overnight incubation at 28°C, a few drops of a 1% solution of N, N-dimethyl-p-phenylene-diamine monohydrochloride were added to the growth on the nutrient agar slant to determine the oxidase reaction.

Aeromonas hydrophila species was/were identified into A.hydrophila, A.sobria and A.caviae group (PALUMBO, et al. 1985 a) and differentiated by their glucose fermentation and esculin hydrolysis reactions and according to OKREND, et al. (1987).

RESULTS

The recovery and differentiation of Aeromonas hydrophila from retail frozen burger, minced meat and sausage are given in Tables (1 & 2).

DISCUSSION

Increasing recovery of A.hydrophila as a food-borne and human pathogen was detected. In recent years, investigations into the cause of human gastroenteritis have resulted in increased concern about A.hydrophila as a possible cause of diarrheal disease in man. At the same time, the role of A.hydrophila as a food-borne pathogen is not fully understood, wherease present information suggests that because of its ubiquitous nature and psychrotrophic characteristic, this organism is a common contaminant on numerous foods (FAGHRI, et al. 1984 and HOOD, et al. 1984).

The present results demonstrated that all examined burger, minced meat and sausage samples were harboured A.hydrophila with average of $30x10^7$, $21x10^7$ and $29x10^7$ respectively. The obtained results are higher than the results reported by OKREND, et al. (1987) who recorded that the indicated numbers of Aeromonas in 10 examined ground beef samples were ranged from 4.44x10 to 7 $4.44x10^3$/g, and appeared to be slightly higher than the obtained results by PALUMBO, et al. (1985 a) who found that the count of A.hydrophila in sausage, ground beef, ground veal and ground lamb was $2.5x10^2$, $5.0x10^5$, $2.1x10^5$ and $2.4x10^6$ after 7 days at 5°C respectively.

Concerning classification of A.hydrophila, the present results are lower than those reported by OKREND, et al. (1987) who isolated A.hydrophial from all 10 ground beef

samples; A.sobria, from four samples and A.caviae from six samples; higher than the results recorded by TERNSTROM and MOLIN (1987) who isolated A.hydrophila from 27% out of 45 examined raw beef samples.

Most studies into sources responsible for A.hydrophila gastro-enteritis have concentrated on its transmission in water supplies (RIPPEY and CABELLI, 1979) but BUCHANAN (1984) alternatevely suggested that the species may present a significant "new" food-borne pathogen and hypothesized that foods may be important in the dissemination of microorganism.

The public health significance of Aeromonas hydrophila in frozen meat products must still be evaluated.

REFERENCES

Alur, M.D.; Lewis, N.F. and Kumta, U.S. (1971): Spoilage potential of predominant organisms and radiation survivors in fishery products. Ind. J. Exptl. Biol., 9: 48.

Buchanan, R.L. (1984): The "new" pathogens: An update of selected examples. Assoc. Food Drug off. Q. Bull., 48: 142-155.

Buchanan, R.L. and Palumbo, S.A. (1985): Aeromonas hydrophila and Aeromonas sobria as potential food poisoning species. A review. J. Food Safety. 7: 15.

Burke, V.; Gracey, M.; Robinson, J.; Peck, D.; Beaman, J. and Bundell, C. (1983): The microbiology of childhood gastroenteritis: Aeromonas species and other infective agents. J. Infect. Dis., 148: 68.

Enfores, S.O.; Molin, G. and Ternstrom, A. (1979): Effect of packaging under carbon dioxide, nitrogen or air on the microbial flora of pork stored at 4°C. J. Appl. Bacteriol., 47: 197-208.

Faghri, M.A.; Pennington, L.C.; Cronholm, S.L. and Atlas, M.R. (1984): Bacteria associated with crabs from cold waters with emphasis on the occurrence of potential human pathogens. Appl. Environ. Microbiol., 47: 1054-1061.

Gracey, M.; Burke, V. and Robinson, J. (1982): Aeromonas associated gastroenteritis. Lancet ii: 1304-1306.

Hazen, T.C.; Fliermans, B.C.; Hirsch, P.R. and Esch, W.G. (1978): Prevalence and distribution of Aeromonas hydrophila in the United States. Appl. Environ. Microbiol., 36: 731-738.

Hood, M.A.; Baker, M.R. and Singleton, L.F. (1984): Effect of processing and storing oysters meats on concentrations of indicator bacteria, vibrios and Aeromonas hydrophila. J. Food Prot., 47: 598.

Lerke, P.; Admas, R. and Farber, L. (1965): Bacteriology of spoilage of fish muscle III. Characterization of spoilers. Appl. Microbiol., 13: 625.

Littel, K.J.; Pikelis, S. and Spurgash, A. (1986): Bioluminescent ATP assay for the rapid estimation of microbial numbers in fresh meat. J. Food Prot., 49: 18-22.

Ljungh, A. and Wadstrom, T. (1983): Toxins of Vibrio parahaemolyticus and Aeromonas hydrophila. J. Toxicol. Toxin. Rev., 1: 257.

Okrend, J.G.A.; Rose, E.B. and Bennett, B. (1987): Incidence and toxigenicity of Aeromonas species in retail poultry, beef and prok. J. Food Prot., 50: 509-513.

Palumbo, S.A.; Maxino, F.; Williams, A.C.; Buchanan, R.L. and Thayer, D.W. (1985 a): Starch-ampicillin agar for the quantitative detection of Aeromonas hydrophila. Appl. Environ. Microbiol., 50: 1027.

Palumbo, S.A.; Morgan, D.R. and Buchanan, R.L. (1985 b): Inflence of temperature, NaCl and pH on the growth of Aeromonas hydrophila. J. Food Sci., 50: 1417.

Rippey, S.R. and Cabelli, J.V. (1979): Membrane filter procedure for enumeration of Aeromonas hydrophial in fresh waters. Appl. Environ. Microbiol., 38: 108-113.

Stern, J.N.; Drazek, S.E. and Joseph, W.S. (1987): Low incidence of Aeromonas sp. in Livestock Feces. J. Food Prot., 50: 1.

Ternstrom, A. and Molin, G. (1987): Incidence of potential pathogens on raw pork, beef and chicken in Sweden, with special reference to erysipelothrix fhusiopathiae. J. Food Prot., 50: 2, 141-146.

Trust, T.J. and Chipman, C.D. (1979): Clinical involvement of Aeromonas hydrophila. Can. Med. Assoc. J., 120: 942-947.

Table (1) : Incidonce of Aeromonas hydrophila recovered from frozen meat products.

Type of samples	No. of samples examined	Positive samples		Count		
		No.	%	Min.	Max.	Average
Burger	15	15	100	27×10^3	20×10^8	30×10^7
Minced meat	15	15	100	24×10^3	53×10^7	21×10^7
Sausage	15	15	100	17×10^7	45×10^7	29×10^7

Table (2) : Differentiation of Aeromonas hydrophila isolated from frozed meat products.

Type of samples	No. of samples examined	Classification of isolated spp./%		
		A. hydrophila	A. sobria	A. caviae
Burger	15	5/33.33	6/40	4/26.67
Minced meat	15	7/46.66	4/26.67	4/26.67
Sausage	15	4/26.67	10/66.67	1/6.66