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MICROBIOLOGICAL QUALITY OF FROZEN LIVER IN ASSIUT
(with 2 Tables)

By

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دراسات ميكروبيولوجية عن الكبد المجمد بأسيوط

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تم فحص ٥٠ عينة من الكبد المجمد والتي تم جمعها من محلات أسيوط المختلفة. ولقد تم تقدير العدد الكلي للميكروبات الهوائية والميكروبات المعوية والميكروب المكور العنقودي الذهبي والتي تراوحت مابين ٢×١٠^{٣} الى ٢×١٠^{٦}/جم و ٢×١٠^{٣} الىٰ ٢×١٠^{٥} /جم و ٢×١٠^{٢} الىٰ ٢×١٠^{٤} /جم على التوالي والمتوسط العددى لهم : ٢٥ر٥ ×١٠^{٥} جم ، ٦٧ر١×١٠^{٤} /جم ، ٧٢ر٦ ×١٠^{٣} جم علىٰ التوالي . وقد أمكن عزل الميكروبات الآتي:Citrobacter spp., Hafnia alvei, Enterobacter spp. Klebsiella spp. and Proteus spp. وقد تم مناقشة الاهمية الصحية ومدى خطورة هذه الميكروبات على صحة المستهلك.

SUMMARY

Fifty random samples of frozen livers were aseptically collected from different shops and supermarkets in Assiut city. The samples were examined for: aerobic plate counts, enterobacteriaceae and staph. aureus counts and for detection of Salmonellae and Shigellae. The aerobic plate count ranged from $2X10^{3}$ to $2X10^{6}$ /g with a mean value of $5.25X10^{5}$ /g. The counts of Enterobacteriaceae and Staph. aureus ranged from $2X10^{2}$ to $2X10^{4}$ /g and $2X10^{2}$ to $2X10^{3}$/g with a mean value of

1.67X10^{3} and 6.72X10^{2} /g respectively. The Enterobacteriaceae which could be isolated from the examined frozen livers samples were identified as: Citrobacter spp., Hafnia alvei, Enterobacter spp., Klebsiella spp. and Proteus spp. Salmonella and Shigella could not be detected in the examined liver samples.

INTRODUCTION

Numberous reports are available in the research literature which describe the numbers and types of bacteria on fresh meats. Most of these reports deal with the microbial flora of surface and interior parts of muscles of beef, pork and lamb carcasses, only a few reports related to the microbial flora of organs, such as liver, Kidneys and hearts (HANNA et al., 1982).

Microbiological contamination on porcine livers is predominantly on the surface and freezing does not change the spoilage characteristics. Beef livers were found organoleptically unacceptable after 7–10 days of storage at 5 °C. This was probably due to a souring type spoilage and bacterial levels of 7.2–7.8X10 organism/g (ROTHENBERG et al., 1982).

SHELEF (1975) reported that the initial counts of beef livers from supermarkets were approximately 10^{5} /g. The microbial flora consisted of Gram–positive cocci, spore formers, coliform bacteria and Gram–negative rods. Following storage in air for 7–10 days at 5 °C, the liver became organoleptically unacceptable due to souring, at which counts had reached 7–8X10^{7} /g. Lactic acid bacteria were predominant in the spoiled livers.

MATERIAL and METHODS

Collection of samples:

Fifty random samples of frozen livers were collected from different shops and supermarkets in Assiut City. All samples were aseptically packageol and brought to the laboratory with minimum of delay.

Preparation of samples:

25 gm of each samples were added to 225 ml of sterile 0.1% peptone water in a sterile blender. The samples was blended for 3 min. at high speed. Serial dilutions from 10 to 10^{-7} were made and then the bacteriological analysis were performed.

Bacteriological analysis:

Aerobic plate count: Standard plate count agar was used for the aerobic plate count according to A.P.H.A. (1972).

Enterobacteriaceae count:

0.1 ml of each dilution was plated on violet red bile glucose agar (VRBG) According to MERCURI and COX (1979). the plates were incubated at 37 °C for 18–24 hr. All purplish–red colonies surround by a red zone of precipitated bile acids were counted. Biochemical tests done on the isolate colonies according to EDWARD and EWING (1972).

Enumeration of coagulase positive staphylococci:

By a surface plating technique, 0.1 from each of the previously prepared dilution was transferred and evenly spread over a dry surface of Baird Parker medium plates (THATCHER and CLARK, 1975). Inoculated plates were incubated at 37 °C for 48 hr. Suspected colonies are counted. Coagulase test was carried out according to CRUICKSHANK et al. (1975).

Detection of Salmonella and Shigella organisms:

10 gm portion of each sample were inoculated into 200 ml selenite cystine broth and incubated at 36 °C for 18–24 hr. A loopfull from incubated broth was streaked on SS agar (Difco). Suspected Salmonella or Shigella colonies were further identified biochemically and serologically according to CRUICKSHANK et al. (1980).

RESULTS

The obtained results are recorded in Table 1 and 2.

DISCUSSION

Aerobic plate count:

The aerobic plate count (Table 1) ranged from $2X10^3$ to $2X10^6$ /g, with a mean value of $5.25X10^5$ /g. These findings agreed with that reported by MATES (1983); TIWARI & KADIS (1981) and HALL et al. (1967). While lower findings were reported by HANNA et al. (1982).

HANNA et al. (1982) reported that freezing can cause sublethal injury and death to many microbial species in foods. Freezing of livers, kidneys and hearts did not cause significant changes in APC.

The count of Enterobacteriaceae in frozen liver samples ranged from $2X10^2$ to $2X10^4$ /g, with a mean value of $1.67X10^3$ /g. The count of Enterobacteriaceae in frozen liver samples showed a close agreement with

that reported by ROTHENBERG et al. (1982); MURTHY (1984) and MADDEN et al.(1986). Types of Enterobacteriaceae could be isolated with different percentage from the examined frozen liver samples as follow: Citrobacter spp. 36%, Hafnia alvei 24%, Enterobacter spp. 16%, Klebsiella spp. 12% and Proteus spp. 12% (Table 2).

ROTHENBERG et al. (1982) reported that Enterobacteriaceae counts for livers in all packaging treatments were low. There were initially very few Staph.aureus organisms, however significantly higher countws were found following both abuse and control treatments. Contamination from the handlers during packaging of the samples combined with the moderate insensitivity to freezing of Staphylococcus, could possibly account for significantly higher numbers following the abuse and frozen storage (control) treatments.

Coagulase-positive Staph.aureus count on Baird-Barker media were ranged from 2X10 to 2X10 /g, with a mean value of 6.72X10 /g. These results are in accordance with the findings obtained by FRUIN et al. (1978) and MATES (1983).

Coagulase-positive Staph.aureus organisms were detected in 2(4%) of the examined frozen liver samples.

Salmonella and Shigella could not be detected in the examined samples of frozen liver.

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Table (1): Aerobic plate count, Enterobacteriaceae and Staph.aureus counts in examined frozen liver samples.

	Minimum	Maximum	Mean
Aerobic plate count	$2x10^3$	$2x10^6$	$5.25x10^5$
Enterobacteriaceae count	$2x10^2$	$2x10^4$	$1.67x10^3$
Staph.aureus count	$2x10^2$	$2x10^3$	$6.72x10^2$

Table (2): Frequency of Enterobacter organisms detected in 50 specimens of frozen liver (125 isolates).

Organisms	No. of isolates	Percentage
Citrobacter	45	36%
Hafnia alvei	30	24%
Enterobacter spp.	20	16%
Klebsiella spp.	15	16%
Proteus	15	12%
Total	125	100%