

DETECTION OF SALMONELLAE IN SOME MEAT PRODUCTS

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ABSTRACT

A total of 150 samples of meat products (34 sausage, 35 beef burger, 39 minced meat and 42 luncheon samples) were collected randomly from different markets at Kafr-El-sheikh and El-Gharbiah governorates on Egypt .the samples were examined for isolation and identification of Salmonella Spp.. The PCR results showed that salmonella Spp were detected in 4 (11.8 %), 1(2.9%), 2 (5.1%) samples of examined sausage, beef burger and minced meat samples, respectively and not detected in luncheon samples. The isolated salmonellae were S. kentucky, S.saintpaul and S.tounouma in sausage samples, S.liverpool in beef burger samples and S. muenster and S. hadar in minced meat samples. These results showed that sausage, beef burger and minced meat might represent a source for salmonella as a food borne disease for human being.

INTRODUCTION

Meat and meat products are high in moisture, nitrogenous compounds, minerals, growth factors, fermentable carbohydrates (glycogen) and of favorable pH, that provides a suitable environment for proliferation of meat spoilage microorganisms and common food-borne pathogens (*Aymerich et al., 2008*). Processed meats are the result of the need to preserve meat in ancient times. The knowledge to preserve meat by making fermented sausages was already known in ancient times (*Pederson, 1980*).

Food borne disease and microbial spoilage of food result from the failure or inability to control microorganisms at one or more stages of the food chain from raw material production to consumption of the final product (*Afshin et al., 2011*).

Food borne infections and illnesses is a major international health problem with consequent economic reduction. It is a major cause of illness and death worldwide (*Adak et al., 2005*).

Salmonellosis is one of the major food borne diseases. Due to its endemic nature, high morbidity and association with a wide range of foods, this zoonotic disease is of high public health concern (*Kottwitz et al., 2008*). There are several transmission routes for salmonellosis, but the major route of human infections is the consumption of contaminated foods especially those of animal origin (*Hernandez et al., 2005*). The public health hazard of salmonella food poisoning in causing self-limiting gastroenteritis characterized by diarrhoea, abdominal cramps and sometimes vomiting and fever and also Typhoid and paratyphoid fevers (*Rhoades et al., 2009*).

Therefore the aim of the present study was to throw the light on the incidence, isolation and identification of Salmonellae from some meat products.

MATERIAL AND METHODS

1- Collection of samples

A total of 150 random samples of meat products (34 sausage, 35 beef burger, 39 minced meat and 42 luncheon samples) were collected randomly from different markets at Kafr-El-sheikh and El-Gharbiah

governorates. The collected samples were transferred directly to the laboratory in their packages. The samples were immediately examined bacteriologically for the isolation and identification of salmonellae.

2- Isolation and identification of salmonella:

2.1. Isolation of salmonellae according to (ISO, 2002).

2.2. Identification of salmonella:

2.2.1. Biochemical identification:

The suspected colonies were purified and identified biochemically according to (*Cruickshank et al., 1975*) and (*FDA, 1998*).

2.2.2 PCR confirmation: (according to Ahmed et al., 2007)

2.2.2.1 Bacterial DNA preparation

2.2.2.2 PCR amplification:

Amplification condition step was performed using a pair of primers specific for the species gene fragment of Salmonella for amplification of the extracted DNA. Primers specific for the species gene fragment of Salmonella:

Salmonella forward primer: 5'- ATCGCTGACTTATGCAATCG - 3' and Salmonella reverse primer: 5'- CGGGTTGCGTTATAGGTCTG 3'.

PCR amplification was carried out using 0.5 ml PCR tube, in which 25 µl of PCR mixture were added for each sample (5 µl Taq Master Mix 5X, 1.25 µl from working solution of each primers, 5 µl of extracted DNA and 12.50 µl distilled water).

The amplification was performed in a DNA thermal cycler (Peltier thermal cycler, USA).

Table (1): The PCR amplification cycles condition

Steps	Cycles	Objective	Temperature	Time
First step:(1)	1	Preheating	95 ° C	2 minutes
Second step: (3)	30	Denaturation	95°C	1 minute
		Annealing	57°C	1 minute
		Extension	72 ° C	2 minute
Third step: (1)	1	Final extension	72°C	5 minutes
Fourth step: (1)	1	Holding	4 ° C	99.99 minutes

The amplicons of 204 bp of Salmonella species-specific fragments were visualized by running in 1% agarose gel by using horizontal gel electrophoresis

2.2.3. Serological identification

The confirmed salmonellae were serologically examined in Clinical Microbiology unit of central health laboratory, Ministry of Health, cairo, Egypt.

RESULTS

Table (2): Incidence of salmonellae in the examined meat products samples

Type of examined samples	No. of examined samples	Positive samples	
		No.	%
Sausage	34	4	11.8
Beef burger	35	1	2.9
Minced meat	39	2	5.1
Luncheon	42	0	0

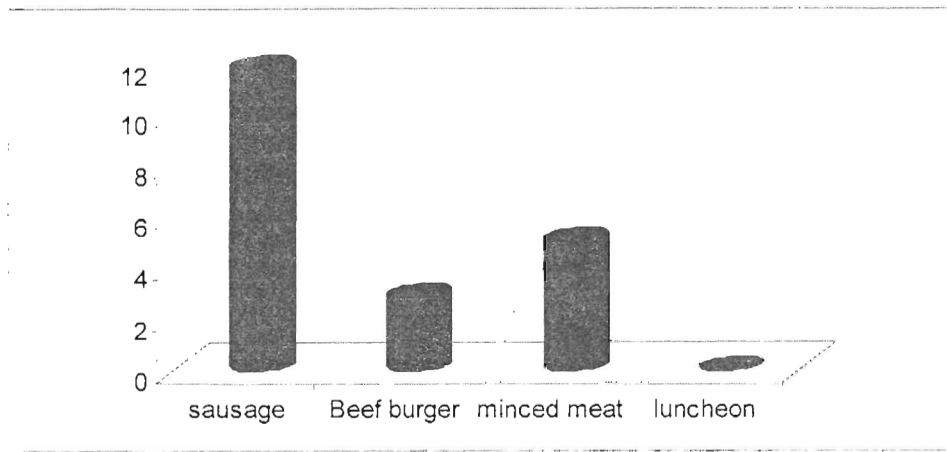


Fig. (1): Incidence of salmonella in the examined meat products samples.

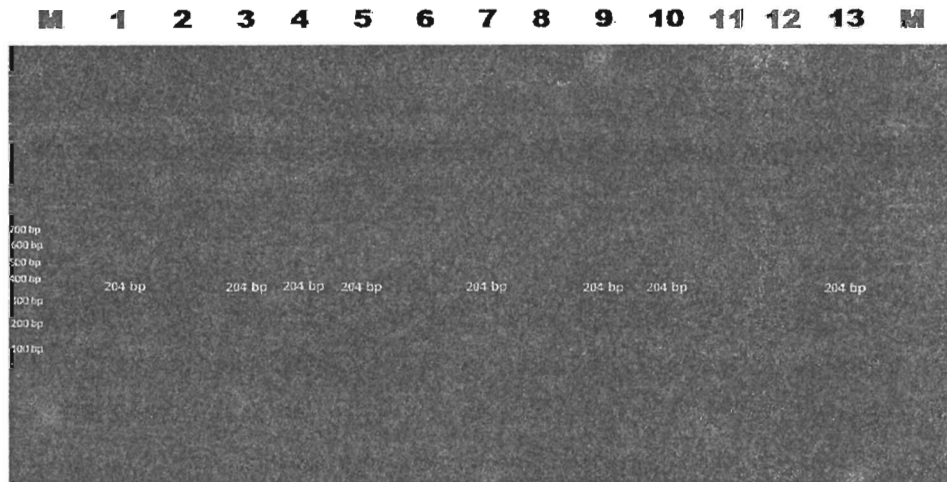


Fig. (2): Agarose gel Electrophoresis of PCR shows that (M= ladder 100 bp, lane 1 control positive, lane 2 control negative, lane 3,4,5,7 the isolated Salmonella spp from sausage samples, lane 6 negative sausage sample, lane 8 negative minced meat sample, lane 9, 10 the isolated Salmonella spp from minced meat samples, lane 11 negative luncheon sample, lane 12 negative beef burger sample, lane 13 the isolated Salmonella spp from beef burger sample.

Table (3): Serotyping of salmonella isolated from the examined meat products samples.

Types of examined samples	No. of Positive samples	+Serovars
Sausage	4	<i>S. Kentucky</i> (2 samples), <i>S.saintpaul</i> and <i>S.tounouma</i>
Beef burger	1	<i>S.liverpool</i>
Minced meat	2	<i>S. muenster</i> and <i>S. hadar</i>
Luncheon	0	0

DISCUSSION

The meat industry is the main reservoir of Salmonella as food borne pathogen which arise from dirt from the hide, as well as dirt of process equipment contaminate carcasses during slaughter and packing house operations (**Braden, 2006**). Consumption of raw or under cooked meat products might cause several food poisoning diseases such as salmonellosis.

It's evident from the results recorded in table (2) and figure 1 and 2 that sausage samples had the higher incidence of Salmonella contamination followed by minced meat then beef burger samples with percentage of 11.8%, 2.9%, 5.1%, respectively. While Salmonellae could not be detected in the examined luncheon samples.

Nearly similar results were obtained by **Eleiwa, (2003)**, **Fath El-Bab-Gehad et al., (2006)** and **Essam, (2010)**, Lower results were obtained by **CORTEZ et al., (2008)**, **Tassew et al., (2010)** and **Sezer et al., (2013)**. Higher results were obtained by **Ejeta et al., (2004)**, **Mrema et al., (2006)** and **Hassanein et al., (2011)** and salmonella could not detected in any samples (**Reham, 2004**, **Malicki and Bruzewicz, 2005** and **Nagwa, 2009**).

The high incidence of Salmonella in sausage may be attributed to the fact that this product is made from raw meat in addition to natural casing is often used in the manufacture which may be important source of Salmonella specially in absence of proper hygiene (*Field et al., 1977*). While in minced meat may be due to cutting and contamination of meat besides the increase in its water and oxygen contents as well as contamination from grinders, air, packaging materials and hands of the workers. Temperature rise (2-4°C) during grinding could also increase the incidence of Salmonella organisms (*Gobran, 1985*).

The presence of even small numbers of M.Os in carcass meat and edible offals may also lead to heavy contamination of minced meat and sausage when meat cutted into pieces, more microorganisms are added to the surfaces of exposed tissue (*Darwish et al., 1986*).

The absence of Salmonellae in luncheon meat may be due to the addition of food additives such as spices and preservatives, which have an antimicrobial activity and inhibit survival and multiplication of microorganisms (*Moffatt et al., 2006*). This also may be due the exposure to high temperature during processing and cooking procedures.

From the result recorded in table (2), it's clear that Salmonella serovars identified from fresh sausage samples were 2 strains as *S. Kentucky*, one strain as *S.saintpaul* and one strain as *S.tounouma* and in the examined beef burger samples one Salmonella serovar was isolated and identified as *S.liverpool*. While in the examined minced meat samples the Salmonella serovars were identified as one strain of *S. muenster* and one strain of *S. hadar*.

From the results of the present study, it could be concluded that *Salmonella* contaminated sausage, beef burger and minced meat samples obtained from retail supermarkets in Kafr-El-Sheikh and El- Gharbia, Egypt. They could be a potential vehicle for food-borne infections and implementation of preventive measures and consumer food safety education efforts are needed. Proper cooking of meat products before consumption and improving personal hygiene in the line of meat products production from farm to processing plant to ensure the safety of meat products for human consumption.

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الكشف عن السالمونيلا في بعض منتجات اللحوم

نظرا لأهميه منتجات اللحوم كمصدر غذائي هام فقد اجريت هذه الدراسه علي بعض منتجات اللحوم في محافظتي كفر الشيخ والغربية وذلك للتعرف علي مدي تلوثها ميكروب السالمونيلا المسبب لبعض الامراض الهامه للانسان مثل حمي التيفويد والنزلات المعويه. لذا فقد تم تجميع عدد 150 عينه ومن منتجات اللحوم 34 عينه سجق، 35 عينه بيف برجر، 39 عينه لحم مفروم و 42 عينه لانشون. وقد أسفرت النتائج عن تواجد السالمونيلا هي 11.8%، 2.9%، 5.1% و 0% في السجق، البيف برجر، اللحم المفروم والانشون علي التوالي. وكانت العترات المعزوله هي *S. Kentucky*، *S.saintpaul* و *S.tounouma* من عينات السجق الطازج و *S.liverpool* من عينات البيف برجر و *S. hadar* و *S. muenster* من عينات اللحم المفروم .

ووقد نوقشت هذه النتائج لمعرفة الاسباب المختلفه لتواجد السالمونيلا بهذه المنتجات والتي أكدت عدم اتباع الاساليب الصحيه السليمه لتصنيع هذه المنتجات وذلك للوصول في النهايه لمنتج صحي امن لضمان سلامه المستهلك.