Bacteriological status of chicken fajitas

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Abstract
A total of random 60 samples of frozen and ready to eat chicken fajitas (30 of each) were collected from different supermarkets and restaurants at El-Monufia Governorates. The collected samples were transferred immediately to the laboratory in an ice box for bacteriological examination which includes the count and study the prevalence of *Staph. aureus* and detection of its enterotoxins, isolation and serological identification of *E. coli* and *Salmonella* in frozen and ready to eat chicken fajitas.

The obtained results revealed that *Staph. aureus* could be detected in six examined samples (20%) and four examined samples (13.33%) with mean value of $4 \times 10^2 \pm 0.43 \times 10^2$ and $3 \times 10^2 \pm 0.18 \times 10^2$ of examined frozen and ready to eat chicken fajitas, respectively.

Isolated *Staph. aureus* were tested for detection of enterotoxins production by using ELISA, the results revealed that two isolates of frozen chicken fajitas were enterotoxigenic, of which one strain was able to produce enterotoxin type A (16.66%) and the other was type D (16.66%) producer, on the other hand one isolate of ready to eat chicken fajitas proved to be enterotoxigenic type (A) with percentage of 25%.

*Salmonella* could be isolated from four examined samples (13.33%) and three examined samples (10%) from frozen and ready to eat chicken fajitas, respectively.

While, *E. coli* could be detected in six examined samples (20%) of both frozen and ready to eat chicken fajitas.

The public health importance and hygienic significance of the isolated strains as well as the suggested measures for improving the quality and safety of the product was discussed.

**Keywords**: chicken fajitas, *Staph. aureus*, *Salmonella*, *E. coli*.

Introduction
During the last decade, the demand of meat and chicken ready to eat products has increased in Egyptian food markets and receive a real consumer preferability because they represent quick easily prepared meat meals and solve the problem of shortage in fresh meat of high price (Reham, 2007).
Chicken meat constitutes an excellent source of high quality, easily prepared and digested protein of the first class which contains all essential amino acids, high proportion of unsaturated fatty acids and less cholesterol, also it characterized by versatility in menu planning (Smith, 2001 and Botsoglou et al., 2003).

Chicken fajitas were manufactured from equal portions of breast and thigh chicken meat from chicken fed either a supplemented level of vitamin E, Rosemary (0.1%), Soya (0.1%) and tea catechins (0.01%) were added to vitamin E (Osullivan et al., 2004).

The processing, handling, distribution and storage as well as marketing of most chicken products may harbor spoilage bacteria responsible for unfavorable changes or constitute a public health hazard due to the presence of pathogenic bacteria leading to harmful effects as food infection or intoxication among consumers (Huvs et al., 2005). Such contamination may render the product of inferior quality or even unfit for human consumption (Baeumler et al., 2000 and Beli et al., 2001).

The presence of a microbial hazard, such as pathogenic bacteria or a microbial toxins, in ready to eat poultry products is one basis on which these products may be harbor the toxins (Levin et al., 2001).

There have been a number of food borne illnesses resulting from the ingestion of contaminated foods such as chicken meat. Most of the pathogens that play a role in food borne diseases have a zoonotic origin (Busani et al., 2006).

Food borne disease has been defined by the World Health Organization (WHO) as a disease of an infectious or toxic nature caused by the consumption of contaminated food (Adams and Moss, 1995).

Staph. aureus food poisoning (SFP) is one of the most common food borne disease resulted from the ingestion of Staph. aureus enterotoxins (SEs) already preformed in food which have superantigenic activity whereas half of them have been proved to be emetic, representing a potential health hazard for consumers (Hennekkinne et al., 2012).

The contamination of food by E.coli of fecal origin were lead to clinical signs include diarrhea, abdominal cramps, nausea, vomiting, chills fever, dizziness within 2-36 hours following ingestion of suspected food (Varnam and Evans, 1991).

Broilers are considerable reservoir for Salmonella infections in man due to the ability of Salmonella to proliferate in the gastrointestinal tract of chickens (Chang, 2000). Salmonellae in man are responsible for a number of clinical symptoms grouped as enteritis and systemic disease; the principle symptoms are fever, abdominal pain, tenderness and diarrhea (Acha and Szyfres, 1991).
Food safety is a global health goal and the food borne diseases take a major crisis on health. Therefore, detection of microbial pathogens in foods is the solution for prevention and recognition of problems related to health and safety \((\text{Velusamy et al., 2010})\).

In Egypt, it is considered that, the main cause of high mortality rate among infants and young children up to 400 deaths and 5 millions illness each year are caused by contaminated meat and poultry products \((\text{El-Seifi et al., 1985 and Gomaa et al., 2002})\).

According to the biological safety, \textit{Staph. aureus, Salmonella} and \textit{E. coli}, are classified into risk group 2 \((\text{Human Pathogen and Toxins, 2009})\) that require containment lab level 2 facilities.

Due to continuous demand for chicken meat products (chicken fajitas) and the increase of consumer’s awareness of the product safety as well as to assure a safe supply, it is extremely necessary not only to increase the production of this important product but also to ensure the bacteriological safety of the product to safeguard consumers against health hazard.

Therefore, the present study was planned out to cover the count and study the prevalence of \textit{Staph. aureus} and detection of its enterotoxins, isolation and serological identification of \textit{E.coli} and \textit{Salmonella} in frozen and ready to eat chicken fajitas.

**Materials and Methods**

1-**Collection of samples:**
A total of 60 random samples of frozen and ready to eat chicken fajitas (30 of each) were collected from supermarkets and restaurants from El- Monufia Governorates. The frozen and ready to eat (sandwiches) chicken fajitas were collected and transferred in an ice box to the laboratory directly without undue delay to be immediately examined for bacteriological examination.

-2- **Preparations of collected samples were carried out according to APHA (1992).**

3- **Enumeration and identification of Staph. aureus were carried out according to FDA (2001).**
3-1- Detection of Staph.aureus enterotoxins by ELISA (Ewalid, 1988): Accurately, RIDASCREEN set C (Art No.: R4101, R-Biopharm AG, Darmstadt, Germany) is an enzyme immunoassay for the detection of Staph.aureus enterotoxins by using their definite kits.

-4-Isolation of E.coli was carried out according to FDA (2002).
-4-1-Serological identification of E.coli according to Kok et al., 1996 by using rapid diagnostic E.coli antisera sets (DENKA SEIKEN Co., Japan).
5-Isolation of Salmonella Spp. was carried out according to ISO 6579 (2002).
-5-1-Serological identification of Salmonella Spp according to Kauffman – White scheme (Kauffman, 1974) for the determination of Somatic (O) and flagellar (H) antigens using Salmonella antiserum (DENKA SEIKEN Co., Japan).

Results and Discussion

Unfortunately, chicken meat products offer an ideal medium for microbial growth as they are highly nutritious, have a favorable pH and are normally lightly salted or not salted at all (Johnston and Tompkin, 1992).

The obtained results as recorded in table (1) revealed that 20% (6 samples) and 13.33% (4 samples) of examined frozen and ready to eat chicken fajitas samples proved to contain Staph. aureus, respectively. Higher incidences were reported by Nahla and Elham (2011), Alvarez-Astorga et al., (2002) and Kreyenschmidt et al., (2003).

Poultry and poultry products ranks first or second in food associated with diseases in most of the countries all over the world which in the USA ranked third of the reported foodborne diseases out breaks (Bean and Griffin, 1990). Epidemiological reports suggest that poultry meat is still the primary cause of human food poisoning. (Mulder, 1999), because the microflora of poultry transferred from the primary production sites to production lines (Fries, 2002).

Also table (1) shows that, the mean value of Staph. aureus were $4 \times 10^2 \pm 0.43 \times 10^2$ and $3 \times 10^2 \pm 0.18 \times 10^2$ of frozen and ready to eat chicken fajitas, respectively. Higher means were recorded by Nahla and Elham (2011), Bailey et al., (2000), Anower et al., (2004) and Eglezos et al., (2008).

Staph. aureus counter considered as an important indicator for the hygienic condition of food (ICMSF, 1996).

The presence of Staph. aureus may be due to contamination of food, equipments. Human and animals are primary resvoirs (Forbes et al., 1998).
The enterotoxins productions of examined samples were mentioned in table (1), the results were revealed that, two samples and one sample of frozen and ready to eat chicken fajitas were harbored enterotoxins, respectively. The enterotoxigenic types were A (16.6%) and D (16.6%) and A (25%) of frozen and ready to eat chicken fajitas, respectively.

The productions of enterotoxins (heat stable toxins) by *Staph. aureus* in food cause nausea, vomiting, abdominal cramps, prostration and diarrhea in human. In more severe cases, headache, muscle cramping and transit changes in blood pressure and pulse rate may occur (Acha and Szyfres, 1991 and Gracey *et al.*, 1999).

The food poisoning with *Staph. aureus* enterotoxins can occur when chicken meat contaminated by large number of bacteria during processing and preserved at temperature higher than 40°C (Jablonski and Bohach, 1997 and Altabari, 1984). Isolation and identification of toxigenic strains of *Staph. aureus* was time consuming and the cultures need to be handled with care because of the zoonotic potential. The biological activity of toxins remains unchanged even after thermal processing of food (Chapaval *et al.*, 2006).

Table (2) shows that, the incidence of *Salmonella* serovars were 13.33% (4 samples) and 10% (3 samples) in frozen and ready to eat chicken fajitas samples, respectively. These results were higher than that recorded by Nahla and Elham (2011), Bucher *et al.*, (2007), Eglezos *et al.*, (2008) and Pointon *et al.*, (2008).

The serotypes of *Salmonella* serovars in frozen chicken fajitas were three strains of *Salmonella enteritidis* group (D1) and one strain was *Salmonella typhimurium* group (B). However the serotypes of *Salmonella* Spp in ready to eat chicken fajitas were two strains of *Salmonella Kentucky group* (C3) and one strain of *Salmonella typhimurium* group (B).

The incidence of *Salmonella* in frozen chicken fajitas were higher than that of ready to eat chicken fajitas as processed raw poultry meat naturally harbors bacteria. Most of these bacteria are responsible for the spoilage of poultry meat. However, poultry products can harbor food borne pathogens from which *Salmonella* Spp and *Staph. aureus* (Wardroup, 1996).

*Salmonellae* is one of the microorganisms mostly associated with outbreaks of illness spread by food. Meat in general and poultry in particular are the commonest source of food poisoning by *Salmonella* (Antunes *et al.*, 2003).

*Salmonellae* are transmitted to man by uncooked meat or by cross contamination from other food that is consumed without further cooking (Gracey *et al.*, 1999).

The clinical symptoms of *Salmonellosis* in man grouped as enteritis and systemic disease. The incubation period of *Salmonella enteritidis* is typically between 6-48 hours. The
principle symptoms are of mild fever, nausea; vomiting, abdominal pain and diarrhea last for few days (1-2 days), in some cases persist for a week or more (3-4 weeks) and

associated with arthritic symptoms. However, the typhoid fever has an incubation period varying from 3-56 days, though it is usually between 10-20 days. There is slow onset of symptoms including fever, headache, abdominal tenderness and constipation followed by diarrhea, peritonitis, septicemia and focal infection at different organs. Fatality rate of typhoid fever 10% compared with less than 1% for most salmonellosis (Ach and Syzfrs, 1991).

Also table (2) recorded that the incidence of E.coli were 20% (6 samples) in each of frozen and ready to eat chicken fajitas.

The serotypes of E.coli in frozen chicken fajitas were three isolates belonged to EHEC (two isolates are O26:H11 and one isolate is O111:H4), two belonged to EPEC (one isolate is O78 and one is O2 : H6), and one belonged to ETEC which is O125 : H21. However in ready to eat chicken fajitas, the serotypes of E.coli were three isolates belonged to EPEC (one isolate is O2:H6 and two isolates are O78) and three isolates belonged to EHEC which are O26: H11.

All the pathogenic strains of E.coli which cause bacterial infection including urinary tract infection , diarrheal disease and other clinical infection such as neonatal meningitis , pneumonia and bacteremia . At least six different categories of pathogenic E.coli causing enteric infections have been identified and further characterized (Alfredo et al., 2010).

The modern poultry processing requires a high rate of throughput to meet consumer demand , as poultry meat can easily be contaminated with microorganisms , due to many factors , as nutrients , high water activity and neutral pH . Also, there are many opportunities for microorganisms to colonize on the surface of the carcasses. Opportunities exist for the contamination of the carcass from the environment, the process in the plant itself, contamination via knives, equipment, the hands of workers and also by cross contamination from carcass to carcass (Kabour, 2011).

The young children , immunocompromised individuals and the elderly are more susceptible to hemorrhagic colitis and show more serious symptoms (Ateba and Mbewe , 2011 and FDA , 2012).

There are other well-known EHEC serotypes that had caused illness world wide, representing a growing public health concern (Lopez-Campos et al., 2012).

Enterotoxigenic E.coli considered as an important cause of diarrheal diseases in enterotoxin (ST). These strains are common cause of travelers diarrhea in many countries (Karmali , 1989 ).
Conclusion and Recommendation
To improve the bacteriological quality of the frozen and ready to eat chicken fajitas and to safeguard consumers from being infected, the hygienic condition of chicken meat product can be assured by the application of hazard analysis critical control point (HACCP) and minimize bacterial proliferation during retailing by storage at -18°C and applying good heat treatment.

Table (1): Prevalence and statistical analytical results of Staph.aureus and its enterotoxins production in the examined chicken fajitas samples.

<table>
<thead>
<tr>
<th>Types of samples</th>
<th>examined samples</th>
<th>Mean±SE.</th>
<th>+ve Staph.aureus No.</th>
<th>%</th>
<th>Enterotoxins Types</th>
<th>No.</th>
<th>%</th>
<th>No.</th>
<th>%</th>
<th>No.</th>
<th>%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Frozen chicken fajitas</td>
<td>30</td>
<td>4x10^2 ± 0.43x10^2</td>
<td>6</td>
<td>20</td>
<td>1</td>
<td>16.6</td>
<td>1</td>
<td>16.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ready to eat chicken fajitas</td>
<td>30</td>
<td>3x10^2 ± 0.18x10^2</td>
<td>4</td>
<td>13.33</td>
<td>1</td>
<td>25</td>
<td>-</td>
<td>0.0</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

N.B: Samples give count <10 cfu/g was calculated as zero.
Table (2) : Incidence and serotypes of *Salmonella* and *E.coli* isolates in examined chicken fajitas samples.

<table>
<thead>
<tr>
<th>Types of samples</th>
<th>No. of examined samples</th>
<th>Salmonella serovars</th>
<th>E.coli serotypes</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>No. of +ve samples</td>
<td>%</td>
</tr>
<tr>
<td>Frozen chicken fajitas</td>
<td>30</td>
<td>4</td>
<td>13.33</td>
</tr>
<tr>
<td>Ready to eat chicken fajitas</td>
<td>30</td>
<td>3</td>
<td>10</td>
</tr>
</tbody>
</table>

References


