كلية الصحة العامة – فلسطين

School of Public Health

القدس – فلسطين

وزارة الصحة

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Deanship of Graduate Studies

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Prevalence of Salmonella in Poultry Meat in Gaza City, 2005

Mahmoud Ahmed Humaid

M.P.H Thesis

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Prevalence of Salmonella in Poultry Meat in Gaza City, 2005

Prepared by

Mahmoud Ahmed Humaid

B.Sc. in Food Science Agricultural Faculty
Zagazeg University- Egypt

A Thesis Submitted in Partial Fulfillment of the Requirements for the Degree of Master of Public Health Al- Quds University

Supervisor

Dr. Yehia Abed

Advisor

Dr. Abed El-jabar El- Tibi

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Al -Quds University
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Thesis Approval

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Prepared By: Mahmoud Ahmed Humaid

Registration No: 20312190

Supervisor: Dr. Yehia Abed, Associate Professor, School of Public Health

Advisor: Dr. Abed El-jabar El- Tibi

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The names and signatures of the examining committee members are as the follows:

1-Head Committee Dr. Yehia Abed Signature----------

2-Internal Examiner Dr. Suzanne Shasha'a Signature----------

3-External Examiner Dr. Abed El-Razek Salama Signature----------

Jerusalem-Palestine

April / 2006

Dedication

To my late father and to my dearest mother, brothers and sisters.

To my beloved wife, and my children.

To my friends.

To my colleagues.

Mahmoud Ahmed Humaid

Declaration

I certify that this thesis submitted for the degree of master is the result of my own research, except where otherwise acknowledged, and that this thesis has not been submitted for a higher degree to any other university or institution.

Signature:

Mahmoud Ahmed Humaid

Date: 01-04-2006

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Mahmoud Ahmed Humaid

- *Definitions:*
- *Bacteria:* ***Living single-cell organisms. Water, wind, insects, plants, animals and humans can carry bacteria, which can thrive on skin, clothes and in human hair, as well as in scabs, scars, the mouth, nose, throat, intestines, and room-temperature foods (PAHO and WHO, 2001[a]).***
- *Carcass:* ***Means the whole of a bird after stunning, bleeding, plucking and eviscerating. However, removal of the kidneys, of the legs at the tarsus, or of the head is optional (Codex, 1976).***
- *Carrier:* ***Person or animal having a specific infectious agent with no clinical signs of disease but capable of transmitting the agent (PAHO and WHO, 2001[b]).***
- *Cleaning:* ***The removal of soil, food residue, dirt, grease or other objectionable matter (Codex, 2003[a]).***
- *Codex Alimentarius Commission****: The Codex Alimentarius Commission was created in 1962 in a Joint FAO and WHO conference about food regulations with the objective of establishing a combined FAO/WHO program based on those regulations. Currently, the Commission has more than 153 member countries that represent almost 97% of the world's population. The Codex Alimentarius create food rules, and guidelines to be followed by the international community as established, with the purpose of protecting consumers' health and ensuring uniform international trade practices (PAHO and WHO, 2001[a]).***
- *Contaminant:* ***Any biological or chemical agent, foreign matter, or other substances not intentionally added to food, which may compromise food safety or suitability (Codex, 2003[a]).***

- *Control Measure:* ***Any action and activity that can be used to prevent or eliminate a food safety hazard or reduce it to an acceptable level (Codex, 2003[a]).***
- *Critical Control Point (CCP):* ***A step at which control can be applied and is essential to prevent or eliminate a food safety hazard or reduce it to an acceptable level (Codex, 2003[a]).***
- *Critical Limit:* ***A criterion which separates acceptability from unacceptability (Codex, 2003[a]).***
- *Cross-Contamination:* ***Is the transmission of a biological, chemical, of physical hazard to a food through dirt, cleaning cloths, contact with other raw products, dirt, or the hands of food handlers (PAHO and WHO, 2001[b]).***
- *Food Handler:* ***Any person who directly handles packaged or unpackaged food, food equipment and utensils, or food contact surfaces and is therefore expected to comply with food hygiene requirements (Codex, 2003[a]).***
- *Food Safety:* ***Assurance that food will not cause harm to the consumer when it is prepared and/or eaten according to its intended use (Codex, 2003[a]).***
- *Food:* ***Any substance, whether processed, semi-processed or raw, which is intended for human consumption, and includes beverages, chewing gum and any substance which has been used in the manufacture, preparation or treatment of "food" but does not include cosmetics or tobacco or substances used only as drugs (PAHO and WHO, 2001[a]).***
- *Food-borne Outbreak:* ***The occurrence of two or more people experiencing the same illness after eating the same food (PAHO and WHO, 2001[a]).***

- *Giblets:* ***Means the liver from which the gall bladder has been removed, the heart with or without the pericardial sac and the gizzard from which the lining and contents have been removed and any other material considered as edible by the consuming country, provided that all such material has been properly trimmed and washed (Codex, 1976).***
- *Good Agricultural Practice in the Use of Pesticides (GAP):* ***Includes the nationally authorized safe uses of pesticides under actual conditions necessary for effective and reliable pest control. It encompasses a range of levels of pesticide applications up to the highest authorized use, applied in a manner that leaves a residue that is the smallest amount practicable. Authorized safe uses are determined at the national level and include nationally registered or recommended uses, which take into account public and occupational health and environmental safety considerations (HACCP). "Actual conditions" include any stage in the production, storage, transport, distribution and processing of food commodities and animal feed (PAHO and WHO, 2001[a]).***
- *Good Manufacturing Practices:* ***Pre-requisites program proceedings, including the, hygienic and sanitary basis needed to implement an adequate HACCP system (PAHO and WHO, 2001[a]).***
- *HACCP:* ***Hazard Analysis Critical Control Points is a system, which identifies, evaluates, and controls hazards, which are significant for food safety (Codex, 2003[a]).***

- *Hazard Analysis:* ***The process of collecting and evaluating information on hazards and conditions leading to their presence to decide which are significant for food safety and therefore should be addressed in the HACCP plan (Codex, 2003[a]).***
- *Hazard:* ***A biological, chemical or physical agent in, or condition of, food with the potential to cause an adverse health effect (Codex, 2003[a]).***

Inspection: Is the examination of food or systems for control of food, raw materials, processing and distribution, including in process and finished product testing, in order to verify that they conform to requirements (Codex, 2003[b]).

- *Inspector:* ***Means a properly trained officer appointed by the controlling authority of a country for the purpose of inspection of meat and meat products and supervision of meat hygiene (Codex, 1985).***
- *Meat:* ***the flesh of animals used as food including the dressed flesh of cattle, swine, sheep, or goats and other edible animals, except fish, poultry, and wild game animals (PAHO and WHO, 2001[a]).***
- *Microorganism:* ***A form of life that can be seen only with a microscope; including bacteria, viruses, yeast, and single-celled animals (PAHO and WHO, 2001[a]).***
- *Pathogen:* ***A microorganism (bacteria, parasites, viruses, or fungi) that is infectious and causes disease (PAHO and WHO, 2001[a]).***
- *Personal Hygiene:* ***Individual cleanliness and habits (PAHO and WHO, 2001[a]).***
- *Poultry:* ***in general means any domesticated bird including chickens, turkeys, ducks, geese, guinea fowls, or pigeons (Codex, 1976).***
- *Prevalence:* ***Number of people suffering from a disease in a specific period of time (PAHO and WHO, 2001[b]).***

- *Preventive Measure:* ***any action or activity that can be used to prevent, eliminate or reduce a hazard to the health of the consumer. Preventive measures refers to sources and factors that interfere with hazards such as the introduction, survival and/or multiplication of biological agents and the introduction and permanence of chemical and physical agents (PAHO and WHO, 2001[a]).***

Risk Analysis: A process consisting of three components: risk assessment, risk management and risk communication (Codex, 2003[b]).

Risk Assessment: A scientifically based process consisting of the following steps: (1) hazard identification; (2) hazard characterization; (3) exposure assessment; and (4) risk characterization (Codex, 2003[b])

- Salmonella*:* ***A bacteria belong to the family of Enterobacteriaceae. Rod-shaped, Gram-negative and non-spore-forming, Main sources of*** **Salmonella** ***are intestinal tracts of domestic animals, and humans (Adams et al 1999, Chin, 2000 and, PAHO and WHO 2001[b]).***
- *Salmonellosis:* ***An illness of humans caused by Salmonellae other than S. Typhi and S. Paratyphi and it is one of most common and widely distributed food-borne diseases. All human pathogens would be regarded as serovars within subspecies S. enterica. All people may contract*** **Salmonella***, but vulnerable groups of the population include infant, young children, elderly and immunosuppressed, where most deaths occurrence in those people (Chin, 2000).*
- *Standard Operating Procedure (SOP):* ***A written method of controlling a practice in accordance with predetermined specifications to obtain a desired outcome (PAHO and WHO, 2001[a]).***

- *Surveillance:* ***The systematic recollection, verification and analysis of data and the dissemination of the information to those who need to know it in order to take actions (PAHO and WHO, 2001[b]).***
- *Virus:* ***A protein-wrapped genetic material that is the smallest and simplest life-form known, such as hepatitis A (PAHO and WHO, 2001[a]).***

Zoonosis: Infection or disease which can be transmitted under natural conditions from vertebrates to man (PAHO and WHO, 2001[b]).

Abstract

- **Salmonella** ***is considered as one of the food-born diseases, and poultry is the main source of*** **Salmonella.** ***This research is a cross sectional design study conducted to identify*** **Salmonella** ***prevalence in fresh, chilled, and frozen poultry (chicken and turkey). It included only worksites licensed by Gaza Municipality that were 32 small-scale places, only one semi automated slaughterhouse in Gaza and two companies dealing with poultry imported from Israel. Data was collected through direct interview and structured questionnaire, prepared by the researcher as well as testing 183 poultry samples. The questionnaire was scrutinized and validated by academic and specialists, and applied on pilot study and samples were examined in Public Health Laboratory of MOH in Gaza.***

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- ***The study showed that fresh, chilled and frozen poultry were contaminated with*** **Salmonella,** ***19.2%, 18.8%, and 0.0% respectively, with a mean average of 16.4%. Fresh, chilled and frozen poultry that had total plate count exceeding level accepted by PS were 2.4%, 21.9%, and 3.8 respectively with average of 5.5%. The study also showed there was no statistically significant relationship between presence of*** **Salmonella** ***and total plate count, but there was a statistically significant relationship between*** **Salmonella** ***and*** **Staphylococcus aureus** ***and*** **E. coli.** ***In addition, it found there was a statistically significant relationship between*** **Salmonella** ***and location of workplaces where Sheikh Rodwan and Shati Camp areas were found to be the highest regarding*** **Salmonella** ***contamination (36.1%). Chicken were of higher contamination(19.1%) than Turkey(3.2%), which reached level statistical significance. The study demonstrated a statistically significant relationship between poultry contaminated with*** **Salmonella, Staphylococcus aureus,** ***or*** **E. coli** ***and type of workplaces where the semi automated slaughterhouse had lower contamination(4.2%) than small scale places (20.7%), but the difference was not a statistically significant with TPC. The study showed there was a statistically significant relationship between*** **Salmonella** ***in poultry and out door environment where workplaces with good out door environment had less*** **Salmonella** ***(5.6%). The study revealed also that there was a statistically significant relationship between*** **Salmonella** ***and methods used for pests control. It showed that places using chemicals for pest control had more contamination (27.7%) than places without any method for pests control (11.4%). Moreover, the study showed there is a statistically significant relationship between*** **Salmonella** ***and type of detergents used in cleaning equipments as liquid detergents reduced contamination. Finally, the study showed no statistical significance relation between*** **Salmonella** ***and worker's knowledge and characteristics or poultry breading places. Places controlled by official organizations MOH and MONE, isolating sick poultry, providing with adequate amounts of water, and selling frozen and chilled poultry had less bacterial contamination.***

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- ***Based upon results, it can be recommended to establish central automated poultry slaughterhouses. As long as that goal will not be achieved in the near future, so it can be recommended to raise awareness of persons dealing with poultry processing for adopting good hygienic practices and improving outdoor***

environment. Imposing and enacting laws and regulations regarding inspection and surveillance of poultry carcasses and other food items for food-born pathogens particularly* Salmonella. *For research purposes, it is recommended to carry out a lager and national wide similar studies to have registered national data about* Salmonella, *its serotypes and its prevalence in food items.

ملخص الدراسة

مدى انتشار السالمونيلا في لحوم الدواجن في مدينة غزة لسنة 2005

تعتبر السالمونيلا من أهم الميكروبات المرضية المنقولة للإنسان بالغذاء كما تعتبر الدواجن أهم مصادرها. هذه دراسة مقطعية أجريت بغرض التعرف على مدى انتشار السالمونيلا في لحوم الدواجن(الدجاج والحبش) الطازجة والمبردة والمجمدة وأحشائها، التي يتم إنتاجها في مرافق ذبح الدواجن سارية الترخيص أو سبق ترخيصها من قبل بلدية غزة سواءاً الصغيرة وعددها 32 والمذبح الكبير الوحيد وكذلك التجار اللذين يقومون باستيراد البضاعة من إسرائيل وتسويقها في قطاع غزة وعددهم 2.

تم جمع المعلومات باستخدام استبيان منظم أعده الباحث وأجرى له التحكيم من قبل أكاديميي ن ومتخصصين بالإضافة إلى تطبيقه علي عينة استطلاعية قبل بدء البحث. شملت الدراسة 183 عينة دواجن من مجتمع الدراسة وتم فحصها في مختبر الصحة العامة الخاص بوزارة الصحة بغزة.

أظهرت نتائج الدراسة أن لحوم الدواجن الطازجة والمبردة وكذلك المجمدة ملوثة بالسالمونيلا بنسبة 19.2% و 18.8% و 0.0% على التوالي وبمتوسط اجمالي قدره 16.4%. كانت نسبة العينات المخالفة لارتفاع العدد الكلي للبكتريا فيها وفقاً للمواصفات الفلسطينية بنسبة 2.4% و 21.9% و 3.8% على التوالي وبمتوسط قدره 5.5%. أفادت الدراسة أنه لا توجد علاقة ذات دلالة إحصائية بين تلوث الدواجن بالسالمونيلا وزيادة العدد الكلي للبكتريا ولكن هناك علاقة ذات دلالة إحصائية إيجابية بين وجود السالمونيلا وعدد بكتريا ستاف أوريس وعدد اشريشيا كولاي.

أوضحت الدراسة وجود علاقة ذات دلالة إحصائية بين وجود السالمونيلا وموقع محلات التجهيز والبيع حيث كانت مناطق الشيخ رضوان والشاطئ أعلى المناطق تلوثًا (36.1%) وكذلك بين تلوث الدواجن بالسالمونيلا ونوع الدواجن حيث كان الدجاج أكثر تلوثا(19.1%) من الحبش(3.2%). أظهرت الدراسة أن هناك علاقة ذات دلالة إحصائية بين وجود ميكروبات السالمونيلا والستاف أوريس و الأشريشيا كولاي ونوعية مذابح الدواجن حيث كان المذبح الآلي أقل تلوثًا(4.2%) من المحلات الصغيرة(20.7%) بينما لا توجد علاقة بين العدد الكلي للبكتريا ونوعية المذابح. أظهرت الدراسة أن العلاقة بين تلوث الدواجن بالسالمونيلا والبيئة خارج محلات الإنتاج ذات دلالة إحصائية حيث كانت الأماكن ذات البيئة الجيدة أقل تلوثاً (5.6%) كما كانت العلاقة بين تلوث الدواجن بالسالمونيلا والإغلاق الجيد للمحلات ضد الآفات ذات دلالة إحصائية حيث الأماكن المغلقة أكثر تلوثاً (37.5%).

أشارت الدراسة إلى أن العلاقة بين تلوث الدواجن بالسالمونيلا وطرق مكافحة الآفات ذات دلالة إحصائية، حيث كانت الأماكن التي تستخدم الكيماويات أكثر تلوثاً (27.7%) من تلك التي لا تستخدم طرق مكافحة(11.4%) أو تستخدم الطرق الميكانيكية والأشعة فوق البنفسجية(6.25%) وكذلك إلى أن استخدام معجون الصابون في التنظيف يزيد التلوث بعكس استخدام الصابون السائل الذي كان فعالا في خفض التلوث.

أظهرت الدراسة عدم وجود علاقة ذات دلالة إحصائية بين تلوث الدواجن بالسالمونيلا وبين الخصائص الشخصية للعاملين من ناحية العمر والتعليم والخبرة والمعرفة وإتباع العادات الصحية أو بين مصدر الدواجن الحية أو بين حالة الدواجن طازجة أو مبردة علماً بأن الدواجن المجمدة كانت خالية من السالمونيلا. أظهرت الدراسة أن المحلات سارية الترخيص كانت أكثر تلوثاً من غيرها ولكن العلاقة لم تكن إحصائية كما كانت المحلات التي يتم مراقبتها بواسطة المؤسسات الرسمية وأخذ عينات منها أقل تلوثاً من غيرها. أظهرت الدراسة أن التخلص من دم الذبح إلى المجاري العامة وتنظيف المعدات مرتين فأكثر يوميًا وكذلك تغير مياه سمط الدواجن أكثر من مرتين يوميا تزيد التلوث ولكن العلاقة غير ذات دلالة إحصائية. أظهرت الدراسة أن عزل الدواجن المريضة وترحيل النفايات الصلبة من المحلات بواسطة أصحابها وبيع الدواجن المبردة والمجمدة وتجميد الدواجن وتوفير المياه بدرجة كافية وإستخدام مواد تطهير للمعدات تقلل من التلوث ولكن العلاقة غير ذات دلالة إحصائية.

أهم توصيات الدراسة تشمل انشاء مذبح آلي لتجهيز الدواجن و حتى يتم ذلك فالتوصية بزيادة وعي العاملين في مجال الأغذية خاصة الدواجن وضرورة الالتزام بإتباع العادات الصحية السليمة وتحسين البيئة داخل وخارج محلات تجهيز الأغذية. كذلك توصي الدراسة بضرورة تفعيل وسن قوانين وتشريعات لسلامة الأغذية و إيجاد نظام للرصد وتقصي الميكروبات الممرضة خاصة السالمونيلا في الأغذية. كما توصي الدراسة بإجراء دراسات بحثية مماثلة على المستوى الوطني للتعرف على مدى وجود الميكروبات الممرضة في الاغذية وبشكل خاص السالمونيلا وأصنافها في الدواجن.

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°C	Degree centigrade (Temperature)
CDC	American - Centres for Disease Control and Prevention
CI	Confidence Interval
Cl	Chlorine
ES	Egyptian Standard
FDA	Food and Drug Administration (USA)
GAP	Good Agricultural Practices
GHP	Good Hygiene Practices
GMP	Good Manufacturing Practices
HACCP	Hazard Analysis Critical Control Point
MOA	Ministry of Agriculture
MOE	Ministry of National Economy
MOH	Ministry of Health
Na Cl	Sodium Chloride
No.	Number
NPHN	National Public Health Network
NRC	National Reference Centre for Salmonella and Shigella
O.R	Odds Ratio
PAHO	Pan American Health Organization
pH	Acidity
PHC	Primary Health Care
PS	Palestinian Standard
PT	Phage Typing
SE	*Salmonella* Enteritidis
ST	*Salmonella* Typhimurium
SPSS	Statistical Package for Social Sciences
USA	United States of America
UV	Ultraviolet

Chapter 1

Introduction

CHAPTER 1

INTRODUCTION

1.1 Background

Food borne infections and intoxications are serious problems for human health. Unfortunately statistical data even from developed countries are scarce and indicate that only a small number of individuals suffer from food poisoning problems every year. Yet available data do not reflect the reality and present only the tip of the iceberg. In case of *Salmonella* it had been estimated that reported cases were only 1:29.5 of the real figure. In 1983, there were 381,881 faecal isolation of *Salmonella* reported to Centre for Disease Control (CDC). If these number multiplied by 29.5 then there would had been 1,146,989 escaping cases (Costanten, 1993).

Anon, 1984 mentioned that poultry was the commonest vehicle that causes *Salmonella* outbreaks. Up to 25% of chicken sold through retail outlets in the UK are contaminated with *Salmonella*, in addition 6% of the outside of retail poultry packaging was contaminated with *Salmonella*. Poultry meat is significant contributors to food-borne illness of *Salmonella* disease. Bean and Griffin, 1990 mentioned that Poultry meat is considered the main source for 90% of food-borne diseases (*Salmonella* 61%, *Staphylococcus* 17%, and *Clostridium* 12%).

Food-borne infections cause an estimated 6.5 million cases of human illness and 9000 deaths annually in the United States. *Salmonella* is the most commonly reported cause of food-borne outbreaks. During 1985-1992, state and territorial health departments reported

437 *Salmonella* Enteritidis outbreaks. Two thirds of patients are less than 20 years of age (CDC [a], 1993). There is no vaccine to prevent salmonellosis (Adams et al., 1999 and Chin, 2000).

Present technology in slaughtering plants cannot guarantee *Salmonella* free final product. Data from 21 countries show median prevalence fresh chicken carcasses of 33.4% (rang 5-73%). The Greece was 59.5%, the prevalence from Poultry reflect the increased chance of cross contamination in the slaughterhouse practices (Costanten, 1998). There are over 21 million cases of typhoid fever reported annually worldwide and 200,000 deaths associated with untreated infection (CDC, 2002).

Dhair and El Husseiny, 2003 shows that in Gaza Strip, there are 108 cases of Salmonellosis infection as reported in epidemiological department during the year 2002. In Gaza Strip during the year 2004, 13out of 80 (16.25%) Chicken samples has been tested were contaminated with *Salmonella* according to Public Health Lab record (Palestine, MOH, 2004[a]).

In Gaza Strip as known, the people buy the chicken after slaughtering and pre cleaning, most of them prepared and cooking at home or restaurants feeding community. The possibility of cross contamination from row poultry to ready to eat food or the row-infected poultry may contaminate the hand of handlers, equipments and other ingredient of the meal. In addition the less awareness in accurate handling of food and personal hygiene may increase the chance of food contamination with *Salmonella*, due to the symptoms of Salmonellosis such as diarrhoea may be not diagnosed as food borne illnesses, also much cases not even go to treatment and not reported. This indicates that there are a high number of infected people with *Salmonella* in Gaza Strip.

1.2 Justification

Annual poultry consumption in Gaza Strip was estimated as 18 Kg. per person in 2005 (Palestine, MOA, 2006). Gaza Strip locally produced about 13.5 million chicken and about 102,400 turkey birds, and imported about 32,900 live turkey birds; also about 3155 tons of frozen poultry meat was imported from Israel slaughterhouses (Palestine, MOA, 2004). The study aims to evaluate the situation of *Salmonella* in poultry meat (fresh, chilled and frozen), due to the role of slaughtering process may be increase the poultry contamination with *Salmonella* in case of not avoids cross contamination, without applying Good Hygiene Practices (GHP) and right process for cleaning and handling. Palestinian Standard (PS) of Fresh Chilled Chicken No. 314/ 1999 banned *Salmonella* species presented in poultry. This agrees with Egyptian standard for chilled poultry No.1651/ 1988, but Saudi standard No.1390/1988 allowed the presence of *Salmonella* in one sample out of five in fresh chilled poultry. The last version of the Egyptian Standard for frozen poultry meat No. 1090/1996 gave the presence of *Salmonella* or absence according to decision of the Minster of Health and Population. Minister of Health and Population in Egypt, in his decision No. 298/ 1980, allowed presence of *Salmonella* in one out of five samples of poultry meat (Ghonaim, 1990).

In France the Centre National d'Etudes et de Recommendations sur la Nutrition et l' Alimentation (CNERNA-CNRS), 1996 specifies for these food products allow presence of *Salmonella* in two out of five samples (in 1 g of neck and skin). Anonymous, 1998 specifies that the Spanish Microbiological Standards allow presence of *Salmonella* in two samples out of five samples (in 10 g of sample). Food Safety and Inspection Service (FSIS) in the USA applied a program for pathogen reduction performance standard for *Salmonella* to measure the effectiveness of the slaughter and grinding process in limiting

Salmonella contamination, and stated not to hold product or recall product based on results of the *Salmonella* samples. Samples taken in sets and the results of the entire set is used to determine if an establishment is meeting the performance standards. So failure to meet *Salmonella* performance standards is based on a set passes, not on individual samples, number of samples for the Broilers is 51 samples, and the maximum number of positives to achieve Standard is 12 samples(20%), *Salmonella* test is positive when any *Salmonella* organisms are found in the unit of the sample which recommended as whole chicken (FSRE, 2004). Capita, et al. 2003 show that contamination of raw meat with *Salmonella* is not generally considered a risk to the consumer because the food expected to heat sufficiently before consumption. Thus eliminating the pathogen although in the majority of countries this microorganism must be absent from ready to eat food products, there are few microbiological norms referring to the contamination of raw poultry products, even though they can cause disease, either directly or indirectly by cross contamination. Therefore, the researcher consider that presence of *Salmonella* in fresh poultry products (raw products) is health and economic problem it need to study to identified clear and taking a rational decision by the relevant decision makers.

1.3 Objectives

General Objective:

To assess the prevalence of *Salmonella* in fresh, chilled and frozen poultry meat.

Specific Objectives:

1- To identify prevalence of *Salmonella* in fresh, chilled and frozen poultry compared to Palestinian Standards.

2- To determine the impact of poultry breading sources, type of slaughterhouses (small or large), and its location on the prevalence of *Salmonella* in poultry meat.

3- To reveal influence of knowledge, practice, and habits of workers on the prevalence of *Salmonella* in poultry meat.

4- To assess the relationship between presence of *Salmonella* and total plate count of bacteria, *Staphylococcus aureus*, and *Escherichia coli* in poultry meat.

1.4 Study Questions

1- Is *Salmonella* present in retail poultry meat in Gaza City and if so by how far?

2- If *Salmonella* is present, is there an influence of handlers on the prevalence of *Salmonella* in poultry meat and if so by how far?

3- If *Salmonella* is present, is there an influence of workplaces on the prevalence of *Salmonella* in poultry meat and if so by how far?

4- If *Salmonella* is present, does locally produced poultry meat differ from imported poultry meat with regard to prevalence of *Salmonella* in poultry meat?

5- If *Salmonella* is present, is there a relationship between its presence and presence of other bacteria considered as indicators of mishandling?

1.5 Study Hypothesis

1- There is a relationship between prevalence of *Salmonella* and handlers and workplaces.

2- There is a relationship between *Salmonella* and other bacteria indicating mishandling.

3- Poultry imported from Israel has less contamination with *Salmonella* than local poultry product.

1.6 Geography and Demography Context

Palestine lies on the Eastern coast of the Mediterranean Sea. It is of an ancient and of strategic important location. Palestinian is compromised of two geographically separated areas namely Gaza Strip and West Bank. Palestinian National Authority (PNA) rules these two parts. Gaza Strip is a narrow zone of land lying along the East Mediterranean Coast and has an area of 360 square kilometers. It is about 50 kilometers long and 5 to 12 kilometers wide and is divided into five governorates that are North Gaza, Gaza City, Mid Zone, Khanyounis and Rafah (Palestine, MOH, 2003). In Gaza Strip, there are five towns, eight refugee camps and fourteen villages (Palestine, MOH, 1999).

Gaza city has an area of about 45 square kilometers. Its main areas include Al- Shajaia, Al-Draj, Al-Toffah, Al-Zaieton, Al-Sbra, Al-Remal, Al- Sh.Rodwan and Al-Shati Camp. Gaza population is assumed to be about 400,000 people; the average national product is estimated at US$ 700 per capita yearly. The city has three universities with 28,500 students (Municipality of Gaza, 2002).

Generally, there are about 4.7 million persons living in Palestinian Territories, of whom 2.3 million (63.2%) live in West Bank and 1.4 million (36.8%) live in Gaza Strip (Palestine, PCBS, 2004; Palestine, MOH, 2004[b]). Demographic reports indicated that Gaza Strip is,

after Hong Kong, the second most densely populated area in the world. The density is about 2,933 people per square kilometer while West Bank is less densely populated with a density of 342 people per square kilometer (World Bank, 1997). About 46.3% of people in Palestine are under 15 years old, and only 2% of population is above 65 years old. In 2004, the average life expectancy in Palestine was 71.1 years for males and 74.1 years for females, and total fertility rate was 4.1 (3.7 in West Bank and 5.5 in Gaza) (Palestine, MOH, 2005[a]).

According to MOH, natural increase of population in Palestine was 2.6% (Palestine, MOH, 2005[a]). But, although population growth rate is decreasing, yet, based on the reported fertility rate at 2003, the population will continue to grow. In addition, Palestinian population is compromised of 51.1% of males and 48.9% of females. Gender predominance toward males below the age of 50 year old, then there is predominance toward females. There is a slight increase in median age for male population in Palestine between 1997 and 2003, where it increased from 16.4 years in 1997 to 16.7 years in 2003 (Palestine, MOH, 2004[b]).

1.7 Socio Economic Context

Employment is the main source of household income and the majority of Palestinian labor force still depends on daily earning of low wages due to the lack of enough jobs in Palestine. Israel still has the upper hand over Palestinian borders, movement and control of goods, so it still holds the economy. The over all adult literacy rate stands at 91% in Palestine, which is higher than the rate of Egypt (56%), but similar to that in Jordan (91%) and less than Kuwait's rate (93%). Over the past 4 decades, male literacy doubled while

female rate increased 8 folds, which indicates no gender gap in this regard (Palestine, PCBS, 2002).

PNA is ranked as middle-income country, with Gross National Production (GNP) per capita of US$ 1,806 in 1999 that decreased to US$ 979 in 2004. Gross Domestic Production (GDP) per capita was US$ 1,496 in 1999, and decreased to US$ 865 in 2004. Number of workers in Israel decreased from 135,000 workers in 1999 to just 50,000 in 2003, and became less in 2004. Workers in Palestine increased from 453,000 in 1999 to 474,000 in 2003 due to the political situation and the current unrest (Palestine, PCBS, 2003).

Despite the economic importance of employment, still there is no reliable data about the actual size of labour force in Palestine. Unemployment is a major socio-economic problem due to constant political unrest that results in frequent closure of borders between the Gaza Strip and the West Bank. Unemployment increased from 11.8% in 1999 to 31% in 2003, due to the political situation and occupations practices including closure of Palestinian regions and cities and other constraints (Palestine, PCBS, 2003). Unemployment causes stress on individuals and families, which leads to an increase of health problems and demands for health care as income is low. In addition, more demand for health care causes overload on health care and allocation of resources to the urgent health services and this leads to deterioration of the health status.

1.8 Layout of Study Chapters

Chapter 1 that is entitled Introduction and it contains background information about retail poultry meat in Gaza and about *Salmonella*. It also contains justification of the conducted study, general and specific objectives of the study, study questions and hypothesis. This

chapter also include some data regarding geography, demography and socio-economic context background in Gaza.

Chapter 2 that is entitled Literature review and it contains information gathered from reviewing related literature regarding characteristic of *Salmonella*, food poisoning by *Salmonella*, economic costs incurred by Salmonellosis, serotypes of *Salmonella* causing disease for humans. It also provides information about *Salmonella* in poultry, methods of reduction *Salmonella* and microbial contamination in poultry meat, other food commodities of animal-origin that can be contaminated with *Salmonella* and poultry feed as a source of *Salmonella* and the control measures to eliminate *Salmonella* during feed preparations. It also includes some research regarding unexpected causes of *Salmonella* outbreaks.

Chapter 3 is entitled Conceptual framework; it contains information about microbiological characteristics of *Salmonella* and situational analysis of retail poultry meat market including live poultry production, marketing and distribution of live poultry and retail poultry meat market in Gaza Strip. It also provides information about factors affecting *Salmonella* prevalence in poultry meat including sources of live poultry, handlers' knowledge and practices, work places locations and intervention policies that affect *Salmonella*. This chapter also include a diagram showing conceptual framework of the study.

Chapter 4 is entitled Methodology; it contains information about study design, inclusion criteria, instrument of the study that included a structured interview questionnaire for workplaces, handlers' characteristics and poultry meat samples tested in Public Health Laboratory of MOH. It also provides information about area of study, setting of study, target population, period of the study, sample size and sampling technique. This chapter include method and instruments used by Laboratory for bacteriological analysis and

procedures. It also provides information regarding pilot study, validation of the instruments, data management, ethical consideration and limitation of the study.

Chapter 5 is entitled Results and contains information about description of results and statistical relationships between the different variables. It includes cross tabulation of various variables and statistical inference regarding handlers' characteristics and knowledge and work places characteristics as sites, having license, outdoor and indoor environment and pest control. It includes cross tabulations of sample results, and description of statistical relationships, against various characteristics of handlers' and workplaces.

Chapter 6 is including Conclusions and Recommendations based upon analysis of results.

Chapter 2

Literature Review

CHAPTER 2

LITERATURE REVIEW

2.1 Characteristic of *Salmonella*

Salmonella belongs to the Enterobacteriaceae family and the genus contains two species: *Salmonella enterica* and *Salmonella bongori*. All of them are motile and have long flagella bacterium except *S. gallinarum* and *S. pullorum*. All ferment glucose but not lactose; all reduce nitrates to nitrites and can survive for several months away from the host (Jordan, 1990). Salmonellae are microscopic living creatures that pass from the faeces of people or animals, to other people or other animals. There are many different kinds of *Salmonella* bacteria. *Salmonella* serotype Typhimurium and *Salmonella* serotype Enteritidis are the most common in the United States. *Salmonella* has been known to cause illness for over 100 years (1885). An American veterinarians scientist named Salmon and Smith, for whom they are named (CDC[b] 1993). The antigenic scheme for classifying Salmonellae recognizes more than 2300 serovares and while all can be considered human pathogens, only about 200 are associated with human illness (D' Aoust, 1997). While Popoff, 2001 mentioned that *Salmonella* according to the O (somatic) antigens, H (flagellar) antigens and Vi (capsular) antigens *Salmonella* strains can be divided into 2501 serotypes. CDC, 2004 revealed that there are 2541 serotypes, some of the serotypes can be further dividing into phage types (PTs) based on host specificity of the bacteriophage.

The reservoir of *Salmonella* are wide range of domestic and wild animals, including poultry, swine, cattle, rodent and pets such as dogs, cats, chicks, also humans. Chronic carriers are rare in humans but prevalent in animals and birds (Chin, 2000). The sources of *Salmonella* contamination are domestic animals, mainly intestinal tract, birds and some reptiles (Idexx, 1998). PAHO and WHO, 2001[a] mentioned that *Salmonella* normally found in the intestinal tract of humans and animals including poultry except for fish, mollusks and crustaceans which can contaminated by these bacteria after being fished. *Salmonella* usually transmitted to humans by eating foods contaminated with animal faeces. Contaminated foods usually look and smell normal, and showed that International Classification of Diseases (ICD) Code of Salmonellosis is 9: 003; 10: A02.0 include infection or food poisoning by *Salmonella* of any serotype, and ICD of Typhoid and paratyphoid fever 9: 002.0; 10 A01.0 while (Palestine, MOH, 2005[b]) showed that the local code salmonellosis is 219 in group B.

Adams et al (1999), Chin (2000) and, PAHO and WHO, 2001[a], described the *Salmonella* germs actually, a group of bacteria with a rod shaped (2-4X0.5µm), non-spore forming, Gram-negative, facultative anaerobic. It can cause abdominal pains, diarrhoea, chills, fever, nausea, vomiting, malaise in humans, and most of its types cause illness for human called Salmonellosis (non-typhoid). The incubation period of Salmonellosis is from six to 72 hours, usually about 12 to 36 hours. The illness usually lasts few days to 7 days, and most persons recover spontaneously without treatment. However, in some persons the diarrhoea may be so severe that the patient needs to be hospitalized. In these patients, the *Salmonella* infection may spread from the intestines to the blood stream, and then to other body sites and can cause death unless the person is treated promptly with antibiotics.

Chin, 2000 mentioned that the dose required causing salmonellosis vary with many factors such as age, general health, nutrition, immune status, whether a person is undergoing medical treatment, and serovar of *Salmonella*. While in general 10^2 - 10^3 cells are required to cause illness. Only a small proportion of cases are recognize clinically, in industrialized countries were there reporting rather than other countries as few as 1% of clinical cases are estimated to be reported. The incidence rate of infection is highest in infants and young children. Most cases occur sporadically (about 60%-80%); elderly, infants, and those with impaired immune systems are more likely to have severe illnesses, which lead to deaths in this group of population.

PAHO and WHO, 2001[a] revealed the control measures and the factors that affecting of *Salmonella* species on handling and the multiplication in food: heating the food to a temperature sufficient to kill bacteria from 65 to 74°C, conserving foods at a temperature below 5°C, preventing cross contamination after cooking, preventing sufferers or carriers of *Salmonella* from working as food handlers, minimum temperature 0°C ± 2°C, maximum temperature 45.6°C, minimum pH 3.7, maximum pH 9.5, the minimum of water activity (A_w)0.945, and maximum Na Cl 8%. FAO and WHO, 2005 declare that freezing usually kills *Salmonella* but some foods e.g. meat appear to be protective of *Salmonella* so freezing does not ensure inactivation. Radiation inactivated *Salmonella* where D value around 0.5 kGy, up to 0.8, but effectiveness depends on food type; D times are higher in drier foods such as desiccated coconut.

The genus *Salmonella* is divided into two species, *Salmonella enterica* and *Salmonella bongori*. *Salmonella enterica* has further divided into six subspecies: *enterica, salamae, arizonae, diarizonae, houtenae* and *indica* (CDC, 2004). Popoff, 2001 revealed that *S. enterica* subsp. *enterica* contains almost all important pathogenic serotypes, the increased

occurrence of *Salmonella* infections within the last decades has accentuated the need for serotyping as a base for proper diagnosis, identification of sources of infection, control of products and to gain better understanding of the global epidemiology of *Salmonella*.

Salmonella are everywhere Gram-negative, facultative intracellular bacteria that replicate in macrophages and neutrophils of the reticuloendothelial systems of numerous animal species, including humans, laboratory and domestic animals, livestock and birds. Several *Salmonella* serotypes including *Salmonella* Typhimurium and *Salmonella* Enteritidis infect a broad spectrum of hosts. Other serotypes such as *Salmonella* Typhi and *Salmonella* Paratyphi in humans, *Salmonella dublin* in cattle and *Salmonella gallinarum* in birds are host specific. In humans, *Salmonella* cause two major types of infection, a systemic disease (typhoid fever) caused by *Salmonella* Typhi, and a gastrointestinal disease (salmonellosis) caused by *Salmonella* Enteritidis or *Salmonella* Typhimurium. Typhoid fever is still a major disease in endemic areas of the world where access to clean water is limited (CDC, 2002).

The genus *Salmonella* contains two species: *Salmonella enterica* and *Salmonella bongori*. Recently, a strain (FRCI) isolated from a low pH, nitrate and U(VI) contaminated subsurface sediment was proposed to represent a new species, *Salmonella subterranea* sp. nov (Shelobolina et al., 2004).

2.2 Food Poisoning by *Salmonella*

Food poisoning by *Salmonella* in hospitals and other health care centres is common and occasionally results in high mortality rates. An outbreak of food poisoning that took place in June 1987 in a hospital in Barcelona, with an attack rate of 59.3%, the mean incubation period was 31.2 hours. The epidemiological evidence pointed to a rice with milk served as

a dessert as the transmitting vehicle, although the contamination source could not be identified. Subsequently, 11 days after the start of the outbreak 9 cases were identified among the patients cared for at the centre, which were considered as secondary cases, the causative organism was *S*E, PT A. The transmission of the mentioned organism within the hospital milieu has not been previously reported (Vaque, et al. 1990).

A human outbreak of *Salmonella* Enteritidis infection occurred following a barbecue in which about 100 persons were involved. Eggs, supplied by one or more of 10 different layer farms, were the most probable source of the infection (Van de Giessen, et al. 1992).

Scott, 1996 said that poultry products is one of the most common causes of food poisoning with Salmonellosis in humans, it caused by the ingestion of *Salmonella* contaminated products. Salmonellosis is the most prevalent food-borne disease in many countries worldwide (D' Aoust, 1997). In 1984, 186 cases of salmonellosis (*S*. Enteritidis) were reported on 29 flights to the United States on a single international airline. An estimated 2,747 passengers were affected overall; food from the first class menu was possibly associated with the disease (FDA 1992). Nagai, et al. 1999 mentioned that an outbreak of *Salmonella* Enteritidis infections occurred in Otaru, Japan, in September 1997. Where there were reported 143 cases of salmonellosis in the local Public Health Centre, in that outbreak one case had a 214-hour incubation period.

In France 1995 the National Public Health Network (NPHN), Ministry of Agriculture (MOA), and the National Reference Centre for *Salmonella* and *Shigella* (NRC) had collected a total of 716 food borne outbreaks that had been reported, they found 108 had been identified as *Salmonella* (Gallay et al., 2000). In a global survey of 104 countries, three serotypes, *S*. Enteritidis, *S*. Typhimurium and *S*. Typhi accounted for 76.1% of all isolates reported in 1995 (Herikstad et al., 2002). There has been a resurgence of

salmonellosis in North America and Europe with an estimated number of salmonellosis of 1.4 million per year in the United States alone (CDC, 2002). In France, the incidence of human salmonellosis recorded by the National Reference Centre for *Salmonella* and Shigella in 2001 was 21 cases per 100,000 inhabitants (Haeghebaert, et al. 2003).

OzFoodNet Working Group, 2003 in Australia mention that there were three outbreaks associated with animal petting zoos or poultry hatching programs and 318 outbreaks of suspected person-to-person transmission. Sites conducted 100 investigations into clusters of gastrointestinal illness where a source could not be identified, including three multi state outbreaks of salmonellosis. OzFoodNet identified important risk factors for food-borne disease infection, including: *Salmonella* infections due to chicken and egg consumption, bakeries as a source of *Salmonella* infection, and problems associated with spit roast meals served by mobile caterers. During three months representing the cool-dry, hot-dry, and rainy seasons of 2002, serological typing was done by the WHO National *Salmonella* and *Shigella* Center in Thailand. Of the food, drinking water, and stool samples from food handlers and healthy persons, 18, 7, 11, and 5%, respectively were positive for *Salmonella* (Vaeteewootacharn, et al. 2005).

2.3 Economic Costs Due to Salmonellosis

Salmonellosis constitutes a major public health burden and represents a significant cost to society in many countries. Very few countries report data on economic cost of the disease, and data related to the cost of Food-borne disease are generally not available from developing countries (WHO, 2005). The annual estimate salmonellosis cost in the Netherlands 225000 $ (Costanten, 1998). The Economic Research Service (ERS), 2003

estimated the total cost associated with *Salmonella* is at US$ 3 billion annually in the United States the estimated medical costs were $0.2 billion, and the estimated value of lost productivity was $2.8 billion, 1.4 million non-typhoid *Salmonella* infections, resulting in 168 000 visits to physicians, 15 000 hospitalizations and 580 deaths annually. Cost estimates per case of human salmonellosis range from approximately US$ 40 to US$ 4.6 million respectively for uncomplicated cases to cases ending with hospitalization and death. In Denmark, the annual estimated cost of food-borne salmonellosis is US$ 15.5 million (2001) representing approximately 0.009% of GDP. A *Salmonella* control programme has been in place for several years in Denmark and the annual estimated cost of this control programme is US$ 14.1 million. It was estimated that this program saves US$ 25.5 million annually of Danish public expenditure. Data related to the cost of food-borne disease is generally not available from developing countries (WHO, 2005).

2.4 Serotypes and Serovars of *Salmonella* and Human Disease.

Eiguer, et al. 1990 show that outbreaks of food-borne diseases are due to *Salmonella* Enteritidis which occurred in Argentina between 1986 and 1988. In 39 registered episodes, 210 strains were isolated from human feces (28 outbreaks) and 59 from food (23 outbreaks). More than 2,500 people in different provinces were affected; the main source of infection was related to raw eggs, eaten in the form of homemade mayonnaise. It is considered necessary to carry out an effective control of poultry products, as well as a permanent surveillance of salmonellosis. Kist, 1991 shows that an increase in human salmonellosis was observed in the Federal Republic of Germany, which was mainly due to an extremely high increase of the incidence of the serovar *Salmonella* Enteritidis (phage type 4), probably reflecting a worldwide epidemiological phenomenon due to the increased

incidence of the serovar Enteritidis, poultry and eggs were identified in various countries as most probable sources and routes of infection.

The spread of *S.* Enteritidis in the Russian Federation occurred mainly at the territories supplied with incubator eggs from the same poultry-breeding enterprise. *S.* Enteritidis strains isolated from infected patients, chicken eggs, follicles and chicken-meat products.

In an outbreak of salmonellosis, registered in 1988 on the Karelian ASSR, 112 persons were affected, one-day old chickens sold to the population by the local poultry plant were the source of infection which was transmitted through everyday contacts; those persons who had direct contact with chickens were affected. *S.* Enteritidis with similar biological characteristics were isolated from salmonellosis patients, from persons having had contacts with chickens and from chickens, chickens were probably infected by oral route (Oblapenko, et al. 1991).

Plasmid analysis of *Salmonella* Enteritidis isolates from human gastroenteritis cases and from two commercial egg-producing poultry flocks determined that the poultry flocks were the source of the human infections with *S.* Enteritidis. Another commercial flock was epidemiologically associated as the source of eggs consumed by affected persons, five *S.* Enteritidis isolates from human cases in these four outbreaks had the same profile and fingerprint, and they all matched those of the 24 isolates from hens in this flock. These results provide further documentation of egg-borne transmission of *S.* Enteritidis to humans (Dorn, et al. 1993).

Glosnicka and Kunikowska, 1994 mentioned that the epidemiological situation in Poland shows that *S.* Enteritidis, in the years 1961-1991, during this period there were two increases in infections and food poisonings, which were of an epidemiological character. The first epidemic, in 1962-1976, affected primarily small children and spread by contact in

a hospital environment. It caused dangerous complications in already existing illnesses, with high mortality, a few foci of later food poisonings were caused by infected meat or meat by-products. The second epidemic which began in 1980 or 1981 and still exists, has already affected about 500,000 persons; it has often concerned cases of sporadic infections, mainly in the case of small children, but it has not been of a hospital epidemic character. It has been far more frequently associated with food poisoning outbreaks caused by contaminated ice-cream, cream cakes, eggs, mayonnaise and, less frequently, by meat and meat by-products. Attention is drawn to the large number of humans transmitting *S.* Enteritidis infections in Poland.

Sparo, et al. 1994 carried out a prospective study From January 1990 to July 1992 on the salmonellosis form of presentation, reservoirs and transmission in Tandil. Forty strains of *Salmonella* enterica were isolated, *Salmonella* Enteritidis was the most frequent isolate, disease was evident only in a few susceptible hosts and the epidemiological chain could not be determined in all the cases. Germany, Austria and Poland experienced 100-200 cases of *S.* Enteritidis salmonellosis per 100 000 people, whereas England peaked at 40 cases and France at 10 cases per 100 000 people (Gomez, et al., 1997).

In poultry phage type 4 was dominant, but in humans, eggs, goats, ducks, sheep, pigs and rabbits, phage type 34 was the dominant type in South Africa, results indicate that phage type 34 was the dominant phage type from 1991-1993, but during 1994-1995 its presence declined. During this latter period the presence of phage type 4 increased. This may suggest that two smaller epidemics consisting of the two different phage types might have been responsible for the epidemic that occurred from 1991-1995 (Mare, et al. 2000).

A prospective case-control study was conducted in Trinidad and Tobago (T&T) to determine the etiology, sources, and risk factors for *Salmonella* Enteritidis (SE) infection.

SE infection in T&T was found to be associated with the consumption of shell eggs, and in particular raw or undercooked eggs. Home-produced eggnog and ice cream, cake batter, and egg-containing beverages were the main raw egg-containing foods, reflecting the cultural practices of the people of T&T. Public health education are needed to reduce the incidence of this infection (Indar-Harrinauth, et al. 2001).

During 1997, *S.* Enteritidis accounted for 85% of all cases of human salmonellosis in Europe, but incidence has declined from this peak (Fisher, 2001). Haeghebaert, et al 2003 mentioned that Salmonellosis is one of the main causes of food-borne infections in industrialised countries. While in France *Salmonella* serotype, Enteritidis represented 39% of the reported cases (Haeghebaert, et al. 2003).

Mason 1994 showed that a sharply rising incidence of salmonellosis in humans caused by *Salmonella* Enteritidis (SE) in the United States between 1985 and 1989 resulted in a government-sponsored outbreak. In addition, Hogue, et al. 1997 show that isolation rate for *Salmonella enterica* serotype Enteritidis (SE) in humans in the United States of America (USA) increased from 1,207 sporadic isolates identified in 1976 (0.6 isolates/100,000 population) to 10,201 identified in 1995 (4/100,000 population). The proportion of reported *Salmonella* isolates which were SE increased from 5% to 25% during the same time period. In 1990, 1994, and 1995, SE was the most commonly reported *Salmonella* serotype in the USA. Although Khazenson, et al. 1996 declare a rise in morbidity caused by *S.* Enteritidis at individual territories of the Russian Federation in the second half of 1980s was due to the consumption of insufficiently heated infected chicken eggs and the non observance of sanitary and hygienic rules in the preparation of food from chicken meat.

A significant increase in the number of isolations of *Salmonella* Enteritidis has been observed in outbreaks of food-borne diseases in humans, associated with the consumption of raw or undercooked hens' eggs. There were 150 outbreaks reported, affecting more than 6000 persons, 71.3% of these outbreaks were confirmed by stool cultures, and 47.3% by bacteriological study of the food implicated in the outbreak (Caffer and Eiguer, 1994).

The dynamics of annual morbidity in salmonellosis caused by *S.* Enteritidis among the population of Perm during the period of 1987-1992 was analyzed, blood sera taken from 4,689 practically healthy donors and from 6,997 hens at poultry breeding. The study revealed that seasonal rises in morbidity caused by *S.* Enteritidis in winter and spring months, as well as in autumn months, were linked with the activation of the epizootic process of *Salmonella* infection among hens at poultry breeding complexes during these periods of the year. A rise in the level of anti *Salmonella* antibodies in poultry and human blood sera was found to be the precursor of the aggravation of the epidemic situation (Sergevnin, et al. 1995).

Boonmar, et al. 1998[a] show a total of 27,497 *Salmonella* isolates from humans, chicken meat, ready to eat Thai foods and shrimps were serotyped to know the predominant serovars of *Salmonella* in humans and foods in Thailand; seventy two and 81 serovars of *Salmonella* were identified in human and food samples, respectively. Sobel, et al. 2000 show that SE cases in the state of Utah increased fivefold. Isolates were identified as phage type 4 (PT4). Forty-three patients with sporadic infections and 86 controls were included in a case-control study of risk factors for infection. A follow up case control study of 25 case and 19 control restaurants patronized by case and control patients examined risks associated with restaurant practices, conclude that SE PT4 transmitted by infected eggs from a single farm caused a fivefold increase in human infections.

Salmonella enterica serovar Enteritidis is the cause of the food-borne salmonellosis pandemic in humans, in part because it has the unique ability to contaminate eggs without causing discernible illness in the birds infected, the infection route to humans involves colonization, survival and multiplication of the pathogen in the hen house environment, the bird and, finally, the egg (Guard-Petter 2001). SE strains were isolated from outbreaks in Brazil, and the most common PT was found to be PT 4, followed by PTs 7, 21, 35, 6, 4a, 8, 30, 6a, 5a, 1, and 1b. Fourteen strains were classified as react but do not conform strains, and one strain was not typeable, the demonstrate PT 4 has a wider distribution among the sources studied than any other SE phage types and is the most important phage type in human salmonellosis (Nunes, et al. 2003).

Salmonella enterica serotype Enteritidis emerged as an important illness during the 1980s, undercooked eggs was the major risk factor for disease, and a variety of prevention and control efforts were initiated during the 1990s, a sporadic infections and outbreaks of S. Enteritidis in the United States from 1985 through 1999. After reaching a high of 3.9 per 100,000 populations in 1995, *S*. Enteritidis infections declined to 1.98 per 100,000 in 1999. While the total number of outbreaks decreased by half, those in the western states tripled. Outbreaks of *S*. Enteritidis phage type 4 infections accounted for 49% of outbreaks in 1999. Outbreak-associated deaths in health facilities decreased from 14 in 1987 to 0 in 1999 (Patrick, et al. 2004).

Salmonellae of non-typhoidal serovars are the most important pathogens involved in food-borne diseases in humans all over the world; the incidence rates of two major *Salmonella* serovars, i.e. *S. enterica* serovar Enteritidis (SE) and *S. enterica* serovar Typhimurium (ST), in the Slovak Republic in 2000-2003 are given. Over the period studied, 829 *S*. Enteritidis strains and 258 *S*. Typhimurium strains isolated from patients with salmonellosis

were investigated in the National Reference Centre for *Salmonella* Phage Typing. The SE strains were differentiated into 16 phage types, with phage type 8 being dominant since found in 73.6%, 53.8%, 62.8% and 45.6% of strains in 2000, 2001, 2002 and 2003, respectively. The following most frequent phage types were 4 and 13a. New phage types, i.e. 15, 5, 25 and 14b, were identified from salmonellosis outbreaks in 2003. The *S.* Typhimurium strains were also differentiated into 16 phage types with phage type DT104 strains being prevalent and showing an increase from 7.4% in 2000 to 44.6% in 2003; the frequency of the other phage types was not epidemiologically significant (Majtanova, 2004).

A comprehensive retrospective analysis of human *Salmonella* Enteritidis isolates in the Bosnia and Herzegovina was conducted in the period 1998-2000. 299 isolates of *Salmonella spp*. were recorded, of which *S.* Enteritidis accounted for 74.2%, the isolation rate of *S.* Enteritidis increased during this period, from 12.7 to 25.5 isolates/year/100,000 population, isolates were obtained all year round, mostly from sporadic cases of infection or limited family outbreaks. Home-made food was identified as the most important source of infection, being implicated in 81% of outbreaks and 81.7% of cases of sporadic infection (Uzunovic-Kamberovic 2004).

Salmonellosis and campylobacteriosis are the most frequently reported acute enteric diseases of infectious origin in the Czech Republic. Morbidity from salmonellosis and campylobacteriosis is highest in the age group 0-4-year-olds. The most frequent causative agents are *Salmonella* Enteritidis (96%), the infection of food-borne from ready-to-eat meals, poultry, confectionery and eggs seem to be most frequently implicated in outbreaks of salmonellosis in public catering and families (Prikazska, et al. 2004).

Non-human sources of *Salmonella* isolates can help identify possible sources of human illness where in USA CDC,2004 reveal that *S.* Typhimurium, the most common serotype in humans, is identified most commonly from clinical samples from bovine sources 2,024 out of 5,359 isolation (38%) while chicken 2.9%, and from non-clinical samples from chicken sources. *S.* Enteritidis and *S.* Heidelberg identified most commonly from clinical and non-clinical chicken sources. However, non-clinical *Salmonella* isolates from non-human sources was 5,676 isolates, the most were from Chicken and Turkey, were the percentage found 35.9% and 32.9% respectively.

CDC, 2004 mentioned that the top 20 most frequently reported *Salmonella* serotypes from Human sources reported which representative 78.1 % of total reporting cases (26,245/ 33,589). *S.* Typhimurium 19.7 %, *S.* Enteritidis 14.5 %, *S.* Newport 11.5 %, *S.* Heidelberg 4.5 %, *S.* Javiana 4.9 %, *S.* Montevideo 2.5 %, *S.* Saintpaul 2.5 %, *S.* Muenchen 2.3 %, *S.* Oranienburg 1.6 %, *S.* Infantis 1.6 %, *S.* Braenderup 1.6 %, *S.* Agona 1.5 %, *S.* Thompson 1.5 %, *S.* I 4,[5],12:i:- 1.5 %, *S.* Mississippi 1.3 %, *S.* Typhi 1.1 %, *S.* Paratyphi B var. L(+) tartrate+ 1%, *S.* Hadar 0.8 %, *S.* Bareilly 0.7 %, *S.* Stanlel 0.7 %.

2.5 *Salmonella* in Poultry

The quality of fresh poultry meat from the microbiological point of view depends mainly on several factors. The first of which is the extent of cleanness of live chicken, and its different preparations in slaughterhouse including slaughtering, hot water-soaking, feathering, intestine- discharging, washing and finally the packaging. Generally speaking, the extent of initial microbial contamination affects negatively on the overall quality of poultry meat (Berrang, et al. 2000 and Buhr, et al. 2000).

Rigby et al. 1980, found the 86.6% of vehicles in which poultry are transported is contaminated with *Salmonella* which mean increasing *Salmonella* number in live poultry that leads to increase contamination during processing. Kampelmacher, 1983 reported that the incidence of *Salmonella* in raw chickens (fresh) were in West Germany (13%) in U.S (45%), in England (35%) and in Netherlands (73%). Safwat et al (1985) isolated *Salmonella* in a rate of (9.05%) in chicken meat and (3.4%) in turkey meat.

Anderson et al (1992) examined 138 samples of poultry gins farm, only 2% of them were positive for *Salmonella*. Hartung, 1993 show that the mean *Salmonella* rate in diagnostic examinations of domestic animals was 5.77%, cattle and chicken showed *Salmonella* rates at the level of the mean rate, while pig, sheep, goats and equines showed much lower levels. On the other hand "other poultry" (9.5%) and especially chicks (13%) had higher levels of *Salmonella*.

Castillo-Ayala, et al., 1993 were isolate *Salmonella* from 69% of fresh chicken and 2.5% of roast chicken; there was no relationship between total plate counts in fresh chicken and isolation of *Salmonella*. Presence of *Salmonella* in chicken is of concern, due to the risk of spreading from the raw food to other cooked foods. The isolation of pathogens from roast chicken indicates mishandling during processing and/or storage of the product. Barrow et al., 1994 shows that in chickens, host specific *Salmonella* such as *Salmonella gallinarum* and *Salmonella pullorum* cause a systemic disease with high mortality rates in birds of all ages.

Wilson, et al. 1996 has been mentioned that the levels of *Salmonella* in retail chickens was about 7%, and *Salmonella* Enteritidis phage type 4 was the most commonly isolated type. The contamination rate is considerably lower than in Great Britain and this may partially explain the lower rate of human *Salmonella* infections in Northern Ireland. Geilhausen, et

al., 1996 show that 1853 packages of fresh chicken breast meat of German, Dutch and French origin were investigated for their contamination with *Campylo bacter* and/or *Salmonella*. *Campylobacter* and *Salmonella* were isolated from 33% and 20% of samples respectively of meat samples. While 6% of these contaminated samples contaminated with both *Salmonella* and *Campylobacter*.

D' Aoust, 1997 mentioned that animal husbandry practices used in the poultry, meat and fish industries and the recycling of offal and inedible raw materially into animal feeds, has favoured the continued prominence of *Salmonella* in the global food chain. Line, et al. 1997 show that the stresses associated with transporting poultry prior to slaughter had been shown to increase pathogen populations both in the intestinal tract and on the carcass exterior before transport, 53.3% of chickens were positive for *Salmonella*. Transport stress increased the colonization rate to 67.5% in control birds, whereas the colonization of yeast-treated chickens decreased to 40%.

Humphrey, et al, 1992; Holt et al., 1998 mentioned that poor ventilation and high dust levels appear to aid dissemination of bacteria among chickens by colonization of mucosal surfaces (i.e. nose and conjunctiva). Dubbert, 1988 determined that poultry contaminated in the process of slaughtering, so it is important to observe the conditions of food cooking.

Boonmar, et al. 1998[b] assessed the prevalence of *Salmonella* in chickens in Thailand in 1997. Twenty-two serovars of *Salmonella* were isolated from 72 of 100 chicken meat samples purchased from 10 retail markets in Bangkok and 20 of 200 chicken meat samples from one slaughterhouse for export and 19 of 285 chicken feces obtained from three farms located in the east region of Thailand. The most predominant serovar was *S.* Enteritidis, which was isolated from 28% of the retail chicken meat, 4.5% of the chicken meat from slaughterhouse, and 6.6% of chickens feces samples examined. Uyttendaele, et al. 1999

analyse 772 samples of poultry carcasses and poultry products for sale on the retail market in Belgium for the presence of *Salmonella* species, *Salmonella* presence in poultry samples was 36.5%. Mulder, 1999; Baeumler et al., 2000 were revealed that Chicken products are widely acknowledged to be a significant reservoir for *Salmonella* and have frequently been incriminated as a source of Salmonellae contamination.

In Gaza Strip, Aljarosha (2001) found that frozen whole meat samples were free of *Salmonella*, while it is positive in three of the 30 fresh poultry meat samples tested which slaughtered locally, and the same result obtained in fresh beef meat. Murakami, et al. 2001 in Western Japan isolate Salmonellae from 37.8% of raw chicken parts. Salmonellae were isolated from faecal samples 34.7% at 35 broiler farms. Salmonellae, including *S.* Enteritidis, were also isolated from swab samples of henhouses associated with one of the shell-egg processing facilities 20%. S. Infantis was dominant in the broiler production environment; sewage samples 61.1% and 22.2% taken from five rivers contained Salmonellae including *S.* Enteritidis (Murakami, et al. 2001).

Zhao, et al. 2001 analysed 825 samples of retail raw meats (chicken, turkey, pork, and beef) for the presence of *Salmonella* serovars and *Escherichia coli* (*E. coli*), the samples were randomly obtained from 59 stores of four supermarket chains during 107 sampling visits in the Greater Washington, D.C., area only 25 (3.0%) of the retail meat samples tested were positive for *Salmonella*. Significant differences in the bacterial contamination rates were observed for the four supermarket chains. Dufrenne, et al. 2001 described contamination levels with *Salmonella* in chicken and chicken products in the Netherlands at retail level using the most probable number method and direct counting, most samples contained <10 *Salmonella* per carcass, both in fresh (89%) and frozen (68%) products.

Beli, et al. 2001 show that 6.5% of 461 chicken meat samples was *Salmonella* positive, taken during a 3-year period, from 1996 to 1998 in Albania. There were no significant differences among years in *Salmonella* positive samples, ranging from 5.1% to 8%, the most frequently encountered serotype was *Salmonella* Enteritidis, isolated in 53.3% *Salmonella* positive samples. but its predomination was clearly evident only in 1996, the other isolated serotypes were *S. senftenberg* (three isolates), *S. newport* (two isolates), and *S. abony, S. agona, S. banana, S. brancaster, S.* Infantis and *S. oslo* with only one isolate each. Four other *Salmonella* strains were not fully serotyped.

Harrison, et al. 2001 analyzed three hundred raw samples (whole chicken, chicken breast with skin or chicken pieces). Campylobacter and *Salmonella* were isolated from 68% and 29% of retail chicken, respectively. *Salmonella* was absent from external packaging but was isolated from 11% of whole packaging. No significant trends in isolation rates of the organisms were obtained during the period of sampling. Chicken and chicken packaging is a potential vehicle for the introduction of pathogens in retail and domestic kitchens and in particular for the cross-contamination of Campylobacter and *Salmonella*.

Wilson, 2002 found that levels of contamination of raw retail chickens with *Salmonella* were 11 %; there was no significant difference between producers contributing large and small numbers of samples. Khosrof Ben Jaafar, et al. 2002 mentioned that 1.7% turkey meat and 3.6% chicken meat, contaminated by *Salmonella*. Dominguez, et al. 2002 were analysed 198 samples of chicken meat for sale in nine provinces of Castilla and Leon (Spain) for the prevalence of *Salmonella*. *Salmonella* was isolated from 71 (35.83%) of the samples analysed. The predominant serovars were *S.* Enteritidis (47.88%), S. hadar (25.35%) and serotype 4,12:b:-(II) (19.71%). Other serovars such as *S.* Mbandaka, *S.* Derby, *S.* Virchow and *S.* Paratyphi B were isolated in much lower levels.

Jorgensen, et al. 2002 show that raw poultry is considered an important source of *Salmonella*, *Salmonella* were present in 25% of the chickens; *Salmonella* were isolated from a sample representing both the inside and outside of the packaging in 19% of the chickens. *Salmonella* was more frequently isolated from samples containing chicken skin in comparison with those containing carcass rinse fluid only, the data presented here contribute to risk assessment and highlight the need to continue to emphasise the safe handling of raw retail poultry.

Salmonella Enteritidis and *Salmonella* Typhimurium infections in young chickens cause also a major disease characterized by severe clinical signs of diarrhoea and dehydration with high mortality rates. In adult chickens, *Salmonella* Typhimurium and *Salmonella* Enteritidis infections do not cause significant disease or mortality and birds can carry the bacteria for several weeks without presenting any clinical signs, which constitutes an insidious risk for public health (Wigley et al., 2002). Salmonellae were detected from 17.9% of the 301 samples examined in Addis Ababa. Chicken meat and giblet samples in 68.2% of the supermarkets were contaminated with Salmonellae; the contamination level of *Salmonella* was higher in chicken giblets as compared to chicken meat, which were respectively 12.3%, 53.1% and 28.0% in chicken meat, gizzard and liver samples (Tibaijuka, et al. 2003).

Capita, et al. 2003 show the average of detection rate occurrence of Salmonellae in retail chicken carcasses and their products in Spain was 49%, the highest was found in chicken carcasses (skin) 55% and the lowest was found in hamburgers 20%. The chicken carcasses purchased in supermarkets were more contaminated which were 75% than those from poultries shops, which were 25%.

Food Standards Agency in UK, 2003 revealed that, a national survey was undertaken in UK, involved testing 4866 samples of fresh, frozen, whole and portioned chicken purchased from over 1500 retail outlets. The overall frequency of *Salmonella* contamination in retail chicken in the UK was 5.7%. There were significant differences between the four countries in the UK. Samples from Wales had the lowest contamination of *Salmonella* (3.4%), with England and Northern Ireland both having a contamination rate of 5.5%, Scotland had the highest frequency of contamination (8.8%). *Salmonella* contamination of fresh chicken (4%) was lower compared to frozen chicken (10.4%) but there was no difference in the frequency of contamination between whole (5.7%) and portioned chicken (5.7%). There was no significant difference in contamination frequency between wrapped and unwrapped chickens or between birds with or without giblets.

The National Reference Centre for *Salmonella* (NRCS) in Austria noticed a cluster of human *Salmonella enterica* subsp. enterica ser. Enteritidis phage type five (*S.* Enteritidis PT5) infections in two neighbouring districts of Austria. Another small outbreak of *S.* Enteritidis PT5 infections that occurred in the same region in 1999 had been traced back to the flocks of a local egg producer (approximately 6 000 hens). Several hundred infections occurred in the course of the 2002 outbreak. By the end of September 2002 the farmer had stopped selling untreated table eggs. In October 2002 only one isolate of *S.* Enteritidis PT5 was ascertained in the region (Berghold, et al. 2003).

Meldrum, et al. 2004 shown that seven hundred thirty-nine samples of raw retail chicken were obtained between November 2001 and December 2002; overall 8% were contaminated with *Salmonella*, there were no significant differences between fresh and frozen carcasses and between samples taken from retailers or butchers. CDC, 2004 shows that isolation *Salmonella* serotype was representative from the poultry (chicken and turkey)

69% (3904/5676) in the non-clinical *Salmonella* isolates from nonhuman sources, chicken 2038, turkey 1866, bovine 220, equine 10, feed/feed supplements 55, other birds /wild animals 78, other domestic animals/ environment 633, porcine 85, reptile 9, all others 682.

Badrinath, et al. 2004 show in epidemiological investigation in United Kingdom strongly suggested the eggs used in the preparation of the egg-fried rice as the vehicle for outbreak. During three months representing the cool-dry, hot-dry, and rainy seasons of 2002, nearly (96-98%) of the fresh pork and chicken, both from the open markets and supermarkets, were positive for *Salmonella* (Vaeteewootacharn, et al. 2005).

Schroeder, et al. 2005 observe from there model suggest that eating *Salmonella enterica* serovar Enteritidis contaminated shell eggs caused 182,060 illnesses in the United States during 2000. Uncertainty about the estimate ranged from 81,535 (5th percentile) to 276,500 illnesses (95th percentile). Our model provides but one approach for estimating food-borne illness and quantifying estimate uncertainty.

2.6 Reduction *Salmonella* and Microbial Contamination in Poultry Meat

Mulder, 1982 mentioned that a radiation of 7KGy leads to a comprehensive annihilation of pathogenic bacteria like *Salmonella*. While Sams, 1994 founded that feed withdrawal 8-10 hours before poultry slaughter but maintaining drinking water, to prevent reduction weight, lead to reduction of the bacteriological count of poultry meat. Drinking water washes the remaining intestinal waste and consequently reduces bacteriological contamination.

Lillard, 1990 mentioned that the microbiological contamination of broilers with aerobic bacteria and *Enterobacteriaceae* has reduced during the preparation, while as *Salmonella* remained without change. However, there was increasing contamination with *Salmonella* during cooling process of poultry meat by soaking in cold water. This is one of the major

cross contaminations of carcass. Where Benedict et al. 1991 found that during the production and preparation, Poultry meat- which is being subjected to changes in temperatures and during soaking in water- stretches and contracts which leads to the detention of bacteria in the cleavages caused by these changes inside the tissues of meat. The removal of detained *Salmonella* inside tissues is greatly difficult because there is some chemical bondings occur between *Salmonella* and bonding tissues such as collagen.

Ishii et al. 1989 Cheng and Beuchat, 1995 Federighi et al. 1995 Capita et al, 2000 found that increasing the accuracy and care in discharging of intestine has reduced the degree of contamination. As well as soaking the carcasses after removing intestine in a solution of NaClO 1-2 ppm for 15 minutes, with repeating washing for several times. Soaking the carcass in a solution of tri-sodium phosphate 10% for 15 seconds, leads to decrease the degree of microbial contamination and inhibition of bacteria stick at the surface of the carcass. Due to the increasing the pH of this solution, there were no changes noted on the sensory properties of both fresh and cooked meat. A reduction of pathogenic bacteria in the wings of poultry meat, such as *Salmonella* and others, has occurred when they treated by a solution of (Lactic acid 0.5%, Sodium Benzoate 0.05%) with a pH 2.64 for 30 minutes.

Lamuka, et al. 1992 and Thayer, et al. 1995 mentioned that the treatment of Poultry meat with Gamma Ray after soaking in different solutions, such as lactic acid, has shown a great reduction of the most bacteriologic contaminants to the degree of complete annihilation for some of them such as *Salmonella*. While treating the poultry meat by the solution alone decreased *Salmonella* to one third of its initial contamination, it is also shown that radiation coupled with solution-soaking increases the shelf life of Fresh broilers by 6 to 15 days when stored at 4°C.

Another method which has been used by Dickens and Whitemore, 1995 to reduce aerobic bacteria and interobacteria, especially *Salmonella* is using a solution of acetic acid 0.6% and cooling for different periods. This reduction becomes more effective by shaking.

Harrison and Harrison, (1996) has been used preheating the meat or poultry jerky strips in the marinade to achieve a minimum internal temperature of 160°F will provide an immediate reduction of *Salmonella*. Morgan et al., 1996 [a, b] have used a modern process to reduce microbial contamination of poultry carcass such as fast pasteurization where the carcass is being subject to very high temperature for a short time under controlled processing by using dry steam at 145°C for 50 minutes, followed by cooling under vacuum and more repeat this step the increase killing. Despite using vapour at 145°C for 25 minute, there was high reduction of pathogenic bacteria. Calicioglu, et al. (2003) indicated that dipping the meat or poultry jerky strips in 5 % acetic acid for 10 minutes before placing it in the marinade could increase the log reduction effects of drying but not enough to eliminate pathogens.

2.7 Other Food Animals' Origin Contaminated with *Salmonella*

Fantasia, et al. 1991 indicated *Salmonella* Enteritidis accounted for 5.45% of the 118.685 *Salmonella* isolates from man and for 2.65% of the 3.315 *Salmonella* isolates from food in Italy in the eleven-year period 1978 to 1988. In the years 1978-1982 no *S.* Enteritidis strain was isolated from eggs and poultry; in the years 1983-1988 the 53% of *S.* Enteritidis isolates from food were from eggs and poultry. In 1989 *S.* Enteritidis accounted for 744 isolates from man and 22 from food of which 80% were from eggs and poultry (partial data). In that year 18 outbreaks caused by *S.* Enteritidis were reported to the National Centre of Enteric Pathogens in Rome.

Epidemiological investigation into an outbreak of food poisoning in 17 patients caused by *Salmonella* Enteritidis phage-type 4 demonstrated a highly significant association with consumption of custard, retailed in custard slices and trifles from a bakery on one day. The bakery had changed their recipe for custard 2 weeks earlier to include fresh shell eggs and had not followed earlier national advice on cooking eggs for human consumption (Barnes and Edwards, 1992).

Grossklaus, 1993 mention that *Salmonella* Enteritidis phage type 4 is mainly responsible to causes illness through eggs or egg-containing food, it has now contaminated the environment including feedstuffs, in addition secondary contamination, occurring during both food production and processing, it is a constant threat to public health. The most important aim must be the creation of *Salmonella* free animal stocks by carrying out regular hygiene control and vaccination programmes and implementing immune prophylactic measures, because of the EC Zoonoses Guideline, redevelopment efforts will have to concentrate in the first place on poultry stocks. It is expected from the EC Egg Regulation, it will drastically diminish the risk of infection from the final product.

Hennessy, et al. 1996 showed that *S*. Enteritidis gastroenteritis developed in 224,000 persons in the United States after they ate Schwan's ice cream, this nationwide outbreak of salmonellosis was most likely the result of contamination of pasteurized ice cream premix during transport in tanker trailers that had previously carried non-pasteurized liquid eggs containing *S*. Enteritidis. To prevent further outbreaks, food products not destined for re-pasteurization should be transported in dedicated containers. Dodhia, et al. 1998 mentioned that home-made ice cream was the vehicle of infection Fresh shell eggs used raw in the ice cream were the likeliest source of infection.

Peresi, et al. 1998 show that there were 906 ill persons including 295 hospitalized patients. Phage type 4 (PT 4) *Salmonella* Enteritidis strains were isolated from 80.5% of stool samples, from all food samples and from 41.7% of eggs, of the outbreaks, 95.7% were associated with the consumption of food containing raw or undercooked eggs. All strains were susceptible to the 13 antimicrobials, except the strains from the nosocomial outbreak.

Seventy-three employees at a food-processing factory employing 2700 staff reported vomiting, diarrhoea, or abdominal pain between 30 July and 3 August 1997. *Salmonella* Enteritidis phage type (PT4) was isolated from 47 symptomatic cases and 5 asymptomatic canteen staff; an uncooked dessert containing raw shell eggs was identified as a possible vehicle of infection (Wilson, et al. 1999).

Nyeleti, et al. 2000 assess the prevalence and distribution of *Salmonella* in the chain from cattle to the consumer, faeces, mesenteric lymphnodes and beef cuts from 235 cattle, stool samples from 300 workers of the abattoir, and 330 minced beef samples from supermarkets in Addis Ababa were analyzed. Low prevalence in faeces and lymphnodes, and higher contamination rates of beef cuts indicate severe cross contamination during slaughter, during transport from slaughterhouse to the supermarkets, production and selling of minced beef, the prevalence of *Salmonella* did not increase.

Outbreak due to the consumption spaghetti which maintained its association (OR = 8.4), the meatballs registered a reduction in risk (OR = 1.8). *S.* Enteritidis was isolated in stool cultures from 28 affected subjects, and in 2 blood and 6 stool cultures from food handlers (5 of whom were classed as cases), *S.* Enteritidis was also isolated in the food samples which content inadequately cooked eggs (Godoy, et al. 2000). In 1995 several outbreaks of food poisoning in humans occurred in Iceland, that were traced to *Salmonella* contamination of singed sheep heads, *Salmonella* infection is rare in Icelandic sheep but

healthy carriers my harbour the bacteria in tonsillae. *Salmonella* was not detected in drainage from slaughterhouses nor in singed sheep heads (Hjartardottir, et al. 2002).

Khosrof Ben Jaafar, et al. 2002 mention that 898 food samples of animal origin analysed to identifying *Salmonella* serovars and to determine the nature of the most contaminated meat products, the results: *Salmonella* contaminate 3.7% of meat product samples. On the 480 samples of bovine meat, *Salmonella* contaminate 4.2%. *Salmonella* contaminate 3.8% ovine meat; *Salmonella* do not contaminate Rabbit meat. Therefore, *Salmonella* contaminates 4.3% of red meat and 2.6% of fowl. *S.* Anatum, Corvallis, Typhimurium, Braenderup, Zanzibar, Enteritidis, Livingstone are different detected serovars. *S.* Anatum represents 40% of contamination whereas. *S.* Typhimurium represents 12% of contaminations.

In the Gaza Strip, Arafa, 2003 found that minced meat contemned with *Salmonella* organisms 14.2 %. Two community outbreaks of salmonellosis that occurred simultaneously in the south west of France, and which was linked to the consumption of fresh Cantal cheese made from raw milk (Haeghebaert, et al. 2003). The source of salmonellosis in human refer to the consumption of contaminated poultry product such as eggs and egg products, 88 % of the cases in Slovakia were caused by *Salmonella* Enteritidis. 228 545 analyzed samples of food and foodstuff of animal origin through 5 years which was examined for *Salmonella spp* the results indicated that 0.21% were confirmed as being *Salmonella* positive and the average ratio of *Salmonella* Enteritidis occurrence in samples was 0.1% per year. A higher incidence (1.43 %) was recorded only in eggs and egg products (Durecko et al. 2004).

2.8 Poultry Feed as a Source of *Salmonella*

Erwin at1955 who the first were discovered *Salmonella* in Poultry feeding, from that time there were care with the feed to control *Salmonella* spread in Poultry production. Kaniawati, 1993 mentioned that the presence of few amounts of *Salmonella* in poultry feeds would reproduce within the live poultry during the first seven days from feeding date, consequently the poultry feeds are one of the main sources causing salmonellosis.

In particular, the ability of *Salmonella* to survive for prolonged periods in dry environments such as feeds has facilitated the potential for recycling of this pathogen through all production stages (Hinton, 1986; Reilly, 1991). The milling of *Salmonella* contaminated feeds and the subsequent risk of oral infection in poultry was demonstrated previously through experimental contamination of feedstuffs and ingestion by young chicks (Williams 1981[a]; Hinton 1988). Henken et al., (1992) used logistic regression analysis on broiler rearing data and concluded that farms supplied with contaminated feeds were 5.3 times more likely to produce *Salmonella* positive flocks, compared to farms which received microbiologically safe feeds.

The reported occurrence of *Salmonella* in poultry feeds and ingredients has varied widely between studies with prevalence ranging from 0% to 78% (Veldman et al., 1995; Bangtrakulnonth et al. 1993; Ward et al., 1996; Macri et al., 1993). Contaminated poultry feeds have been identified as high-risk vehicles for the introduction of infectious agents of public health significance to commercial poultry flocks (Tompkin, 1994; Gowda, 1995; Ha et al., 1998). The prevalence of *Salmonella*, in a dedicated commercial poultry feed mill was undertaken, *Salmonella* was frequently recovered in samples taken in the preheat and post heat treatment areas of the mill with the overall percentage of samples positive found to be 18.8% and 22.6%, respectively. Feed ingredients and dust collected in the preheat

treatment locations within the mill were frequently contaminated with *Salmonella* (11.8% and 33.3% of samples, respectively), 24.2% obtained from the post heat treatment area of the mill and from feed delivery vehicles 57.1% (Whyte et al., 2003).

Control Measures to Eliminate *Salmonella* During Milling Procedures

The control and elimination of *Salmonella* during milling procedures has proved difficult. Since 1991, in Ireland, it has been mandatory that poultry feeds be heat treated to a minimum core temperature of 75°C for a period of one minute or equivalent (Anon, 1991). The heat treatment of feeds has been demonstrated to be an effective means of reducing *Salmonella* in finished feeds. Conventional pelleting technologies used in mills have been shown to reduce levels of Salmonellae in feeds but were unable to eliminate the organism (Williams, 1981 [b]). Whyte et al., 2003 reported that pelleting at 71.1 – 82.2°C for 2 to 16 seconds was insufficient to eliminate *Salmonella* from inoculated feed samples. When the samples were extruded at 93.3 – 176.7°C for 45 to 60 seconds, the organism was no longer detected.

2.9 Unexpected Source of *Salmonella*

Food-borne outbreaks of salmonellosis are usually associated with the consumption of contaminated animal products, or with faecally contaminated fresh produce, but during the last a few years' salmonellosis outbreaks linked to unexpected food sources, these products have a long shelf life, more than 1 year, and are ready to eat foods. In 2001, there was an international outbreak of *Salmonella* Typhimurium DT104 due to consumption of a particular brand of halva, a sweet made from sesame seeds, honey and flavourings, in Germany, Sweden, Norway and Australia. *Salmonella* Typhimurium DT104

was isolated from samples of the halva the patients had consumed (WHO, 2004). OzFoodNet Working Group (2004) mentioned that in 2003, 55 human cases of *Salmonella* Montevideo in Australia linked to the imported tahini from the Middle East, OzFoodNet Working Group (2003) identified the tahini from Egypt, were reported which led to several product recalls.

In the United States, 32 cases Salmonellosis have been identified traced to the consumption of natural raw almonds, with onsets between March 2003 and April 2004. These almonds from California Farms were sold across the United States under several brands and exported to China, Democratic Republic of Korea, France, Italy, Japan, Malaysia, Mexico and the United Kingdom. FDA has notified those countries who received the almonds and several products that contained the implicated almonds have been recalled (WHO, 2004). Isaacs, et al. 2005 show that in Canada, an outbreak from consumed raw almonds where *Salmonella* Enteritidis (SE) phage type 30 (PT30), a rare strain, was detected, in unopened product samples and on farms where the almonds were grown, and they found an association between the patient and almond consumed.

2.10 Summary

It can be assumed that *Salmonella* is rod shaped, non-spore forming, Gram-negative and facultative anaerobic bacteria, belongs to Enterobacteriaceae family and contains two species: *Salmonella enterica* and *Salmonella bongori* and has been known to cause illness for over 100 years. Cosmopolitan, facultative intracellular bacteria have poultry as most important reservoir for human infections. *Salmonella* Typhi and *Salmonella* Paratyphi infect humans only, but other serotypes infect a wide range of domestic and wild animals and bird as well as humans where infection occurs through eating contaminated foods. The

infective dose varies greatly but in general, 10^2 - 10^3 cells are required to cause gastrointestinal illness. *Salmonellae* are killed at temperatures 65 to 74°C. They need pH 3.7 to 9.5, water activity not less than (A_w) 0.945 and NaCl not more than 8%.

Salmonella food poisoning occurs sporadically and mostly affects elderly and infants, but occasionally results in high mortality rates even in hospitals and involving various food items mostly eggs and poultry products. Salmonellosis constitutes a significant cost in many countries that was estimated as US$ 3 billion in the United States resulting in 168,000 visits to physicians, 15,000 hospitalizations and 580 deaths annually. *Salmonella* control programme in Denmark costs reached US$ 14.1 million, but it was estimated to save US$ 25.5 million.

Salmonella in young chickens cause a major disease characterized by diarrhoea and dehydration with high mortality rates. In adult chickens, *Salmonella* do not cause significant disease or mortality, but birds carry the bacteria for several weeks without clinical signs, which constitute an insidious risk for public health. Chicken products are widely acknowledged, and have frequently been incriminated as a source of Salmonellae. Role played by fresh poultry meat in Salmonellosis depends mainly on several factors as cleanness of live chicken, its preparations in slaughterhouse and vehicles transporting poultry.

In some countries, it was found that only 2% of poultry farms, but 69% and up to 72% of fresh chicken, samples were found positive for *Salmonella*. In addition, it was found that 20% of samples from retail market poultry meat were positive. In Gaza Strip, *Salmonella* was detected in 10% of fresh poultry meat samples. It was said husbandry practices used in poultry and in meat industries and incorporating offal and inedible raw material into animal feeds lead to presence of *Salmonella* in global food chain. Poultry can be contaminated

during slaughtering, so it is important to observe its cooking. *Salmonella* was detected in food items other than poultry meat including homemade ice cream and mayonnaise, inadequately cooked eggs**,** minced meat and cheese made from raw milk. Salmonellosis outbreaks were linked to unexpected food sources including halva, honey and flavourings, tahini and raw almonds.

Chapter 3

Conceptual Framework

CHAPTER 3

CONCEPTUAL FRAMEWORK

3.1 Microbiological Characteristics of *Salmonella*

Salmonellae are bacteria belonging to the family of *Enterobacteriaceae*. The genus has two species: *Salmonella enterica* that include six subspecies and *Salmonella bongori* (Adams, et al 1999, Chin, 2000 and, PAHO and WHO 2001[a]). Bacteriologically, *Salmonella* bacteria are rod-shaped, Gram-negative and non-spore-forming. They are motile bacteria with two non-motile exceptions: *S. gallinarum* and *S. pullorum* (FDA, 1992). Main sources of *Salmonella* are intestinal tracts of domestic animals, humans, birds and some reptiles. Fish, mollusks and crustaceans are not original reservoir but can be contaminated by *Salmonella* after being fished (PAHO and WHO 2001[a]). Nowadays, there are 2,541 serotypes based on O and H antigens (CDC, 2004).

Salmonellosis is an illness of humans caused by Salmonellae other than *S.* Typhi and *S.* Paratyphi and it is one of most common and widely distributed food-borne diseases. In many countries, it constitutes a major public health burden and incurs significant costs that include visits to physicians, medicines, hospitalizations, loses of productivity, and may lead to death (WHO, 2005). All human pathogens would be regarded as serovars within subspecies *S. enterica*. Worldwide, millions of human cases of Salmonellosis are reported every year, and the disease leads to thousands of deaths. All people may contract *Salmonella*, but vulnerable groups of the population include infant, young children, elderly and immunosuppressed, where most deaths occurrence in those people (Chin, 2000). In

the Gaza Strip, Central Laboratory of MOH reported 35 *Salmonella* isolations, including 21 positive stool cultures out from 2,398 food handlers (0.88%) in 2005 (Palestine, MOH, 2005[c]).

3.2 Operational Definitions

Case Definition of *Salmonella* in poultry meat:

The positive *Salmonella* case is defined as the presence of *Salmonella* in 25 g poultry sample. Poultry sample. Any part of poultry carcasses either meat or giblets was considered a sample.

Poultry of the Study:

The study was involved with edible chickens (broiler), turkeys' meat, and their giblets.

Slaughter house:

Small scale slaughter houses means shops of small area dealing with poultry slaughtering, processing and selling, in the same time of keeping live poultry in the shops.

Large scale slaughter houses means places of large area dealing with poultry slaughtering and processing using semi automated techniques and selling processed poultry in a separate place.

3.3 Situational Analysis of Retail Poultry Meat Market

3.3.1 Live Poultry Production in Gaza Strip:

It is noticed that poultry are raised in small scale, mostly less than 10,000 birds farms. One-day old chicks raised in these farms are produce exclusively by ten local hatcheries, but the eggs used for hatching are mostly imported from abroad with minor contribution of local parent-farms. Eggs imported for hatching come from USA, Turkey, Spain and Israel

and are under strict supervision of MOA for controlling of *Salmonella* and other egg born diseases of poultry. Veterinary Certificates should accompany imported eggs from exporting countries certifying that eggs came from farms free of *Salmonella* and other egg born causative agents of poultry diseases.

Half of poultry feeds are produced by four local factories in Gaza Strip mostly by Nafco factory, the other amounts received from West Bank and Israel. Farms raising chicken are under supervision of MOA and regular samples are taken from these farms, but only 13 samples were taken in 2004 and all were negative for *Salmonella*, while the number of raising farms are greatly larger that figure (Saed Seyam 27/7/2005, Personal Communication).

3.3.2 Marketing and Distribution of Live Poultry in Gaza Strip:

It is noticed that about 5 relatively large scale companies and many other relatively small scale companies are trading with live poultry. Their work involves purchasing live birds from farmers and selling them to workplaces of slaughtering and processing poultry. It is noticed that they do not clean and disinfect the cages or transporting trucks they use for collection and distribution of live birds. It is expected that such cages and trucks may be contaminated with *Salmonella* from a certain farm and transmit it to poultry of other farms**.**

3.3.3 Retail Poultry Meat Market in Gaza Strip:

Gaza Strip through year 2005 produced about 16.5 million chicks; produce about 18,260 tons of meat, and about 55,000 turkey birds; produce 482 tons of meat. Imports include about 35,000 live turkeys' produce 342 tons of meat, and about 4,243 tons of chilled and frozen chicken and turkey meat that are solely imported from Israel. Poultry individual

consumption in Gaza Strip is about 18 Kg. per year (Palestine, MOA, 2006). Working places licensed by Municipality of Gaza, were 35 in Gaza City, including 32 small scale places that sell freshly processed poultry to consumers, one large scale semi automated place that sell chilled or frozen poultry to all Gaza Governorates and some times to West Bank, and also 2 places that sell chilled and/or frozen poultry imported from Israel. All those places were included in the study, but there are many other places that were not registered or licensed by Municipality therefore they were excluded.

There is no official Veterinary inspection of poultry carcasses in local slaughterhouses, but it is present only for poultry carcasses intended for the hospitals of MOH. It must be mentioned that poultry parts imported from Israel come frozen or chilled, are from licensed slaughterhouse, and certified to receive official inspection by Israeli authorities and it also receives further inspection on arrival to crossing points of Gaza borders, that done by the inspectors of MOA, and MONE.

3.4 Factors Affecting *Salmonella* Prevalence in Poultry Meat

3.4.1 Source of Poultry:

Source of poultry affect on the prevalence of *Salmonella* in live birds can be transmitted to poultry meat through processing. Farmers can reduce *Salmonella* presence in their live birds when applying Good Agriculture Practices (GAP). That includes supervision of hatching egg, design, licensing and control breeding place, pest control and feeding control.

3.4.2 Personal Knowledge and Practices:

These factors are related to knowledge of persons with respect to microorganisms, the role these microorganisms in causing diseases, its reservoir, its ability to contaminate foods, and

methods that destroy them. When people are aware about *Salmonella* as hazardous micro-organisms capable of causing illness, they will try to prevent it by adopting personal hygiene and good hygienic practices, which include:

1. Keeping their hands clean (see annex 10, MOH poster).
2. Wearing special clothes for work and keeps it clean.
3. Exclude the food handlers in case of illness and injuries persons especially diarrhea.
4. Prevent cross contamination during the process of poultry slaughtering.
5. Keeping Chilled poultry out of the dangerous zoon temperature that more than $5^{\circ}C$.
6. Good knowledge of Pest control and preventing its presence in places of food processing and preparation.
7. Good knowledge and practice of adequate cleansing and sanitation of work places and instruments.

3.4.3 Slaughterhouses:

Places that slaughter poultry affect on *Salmonella* presence in poultry meat can contaminated during processing. Slaughterhouses can reduce *Salmonella* presence in their products when applying Good Manufacturing Practices (GMP). That includes supervision live birds; they must be from known and licensed farm, well designed workplaces, licensed and under official control. It should be subjected to internal veterinary inspection, pest control, adopting personal hygiene and good hygienic practices, good cleansing and disinfecting and qualified and trained workers.

3.4.4 Intervention Policies that Affect *Salmonella* Prevalence:

These interventions include imposing and enacting Food Law, regulations and complete set of standards of food safety that must be in harmony with related international standards such as Codex Alimentarius Commission. They also include making agreements to avoid duplications of efforts, responsibilities and interventions of official parties. There are no such applied intervention policies, but some food standards and only minor cooperation between various parties taking part in food safety.

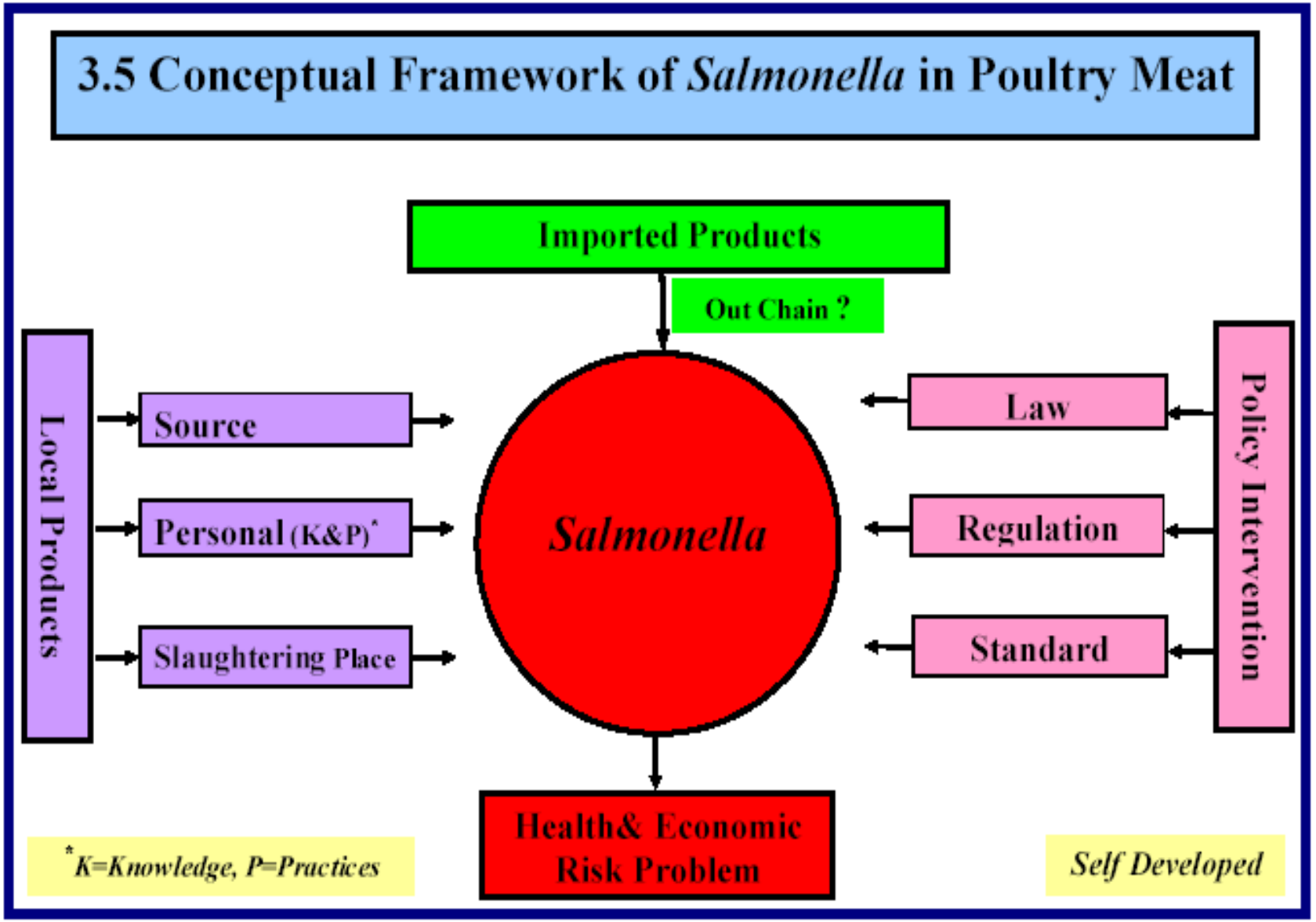
Intervention policies related to official interventions to control diseases and improve environmental safety, it includes obligatory medical examinations of all persons working in food plants particularly poultry processing and sales. It also related to personnel follow up whom are positive for *Salmonella* and preventing them to handling food items till they recover and this, in particular, applies to Preventive Medicine Department of MOH. This policy is only partially present because there are many food handling places that work without license or medical examinations.

It also includes imposing and application of good hygienic and good environmental conditions, official licensing of food handling places and monitoring microbial analysis for foodstuff especially high-risk foods of animal origin. These actions should be carried out by all various official parties taking role in food safety namely MOH, MONE and Gaza Municipality in a cooperative and coordinated manner. Factual situation in Gaza includes this policy but unfortunately, it is not in a proper manner.

Supervision and restriction of imported eggs for hatching and one-day chicks to be comply with MOA requirements that state it should come from farms free of *Salmonella*, as mentioned on annex 11. Also, fresh chilled chicken should be free of *Salmonella* species as mentioned in PS. This policy is adopted and implemented, but number of tests carried out is not enough. This study aimed to know the affect of knowledge and practice of food

handlers. In the main, time the range of effecting source of poultry and the place of slaughtering on the presence of *Salmonella* in poultry meat. In addition, to assess the presence of *Salmonella* in poultry meat that imported out of border from Israeli side, which are unknown histories chains of manufacturing.

According to obtained results from the lab., bacteriological analysis for the poultry samples which taken from the slaughterhouses of poultry in which chicken are mechanically cleaned, chilled, frozen and sold to the supermarkets, retails and consumers, small slaughterhouses which clean and sell fresh poultry for consumers, and suppliers' centres of the chilled and frozen poultry products imported from Israeli slaughterhouses. The recommendation recorded at the end of the study according to the results obtained. Finally, the following diagram reveals the conceptual framework of the study.


3.5 Conceptual Framework of *Salmonella* in Poultry Meat
Imported Products
Out Chain ?
Local Products
Source
Personal (K&P)*
Slaughtering Place
Salmonella
Law
Regulation
Standard
Policy Intervention
Health& Economic Risk Problem
*K=Knowledge, P=Practices
Self Developed

Chapter 4

Methodology

CHAPTER 4

METHODOLOGY

4.1 Study Design

A cross sectional quantitative approach was chosen for this study to assess the prevalence of *Salmonella* in the fresh, chilled and frozen Poultry meat in Gaza City.

4.2 Target Population

Workplaces that slaughtering poultry and selling either fresh, chilled and frozen edible poultry meat, and places that importing chilled and frozen edible poultry meat from Israel, (including persons, place and poultry meat).

4.3 Area of Study

Gaza City was selected as a site for the study where most of providers are present in the city. The researcher considered the selected place as explained in the study setting

4.4 Study Setting

1. Poultry slaughterhouses in which chicken are mechanically cleaned, chilled, frozen and sells to the Supermarkets, retails and consumers.

2. Small slaughterhouses which clean and sell fresh poultry for consumers directly.

3. Suppliers' centres of the chilled and frozen poultry products imported from Israeli Slaughter houses.

4.5 Data Collection Period

- May 2005, Pilot Study.

- September and October 2005.

4.6 Sample Size

The research hypotheses of *Salmonella* in poultry meat are that the maximum prevalence rate about 10% and minimum prevalence rate about 5%, with confidence interval 95%, by using Epi Info programme, the samples was determined by 138. the actually samples were collected from thirty two slaughterhouses where chicken prepared and cleaned; also from the one automatic slaughterhouse, which prepare and sell cleaned chilled or frozen poultry for public and two main supplying centres of the chilled or frozen poultry products, which imported from Israeli slaughterhouses are 183 samples.

4.7 Sampling

The study included all slaughterhouses, which registered in Gaza municipality, and the Suppliers' of chilled and frozen poultry in Gaza city. Include, purposive non-probability sampling.

4.8 Eligibility Criteria

Inclusion Criteria

All licensed or registered workplaces in the Municipality of Gaza City were included.

Exclusion Criteria

Non-licensed or non registered workplaces in the Municipality of Gaza City.

4.9 Data Collection

The researchers collected data through filling structured questionnaire and taking samples. Samples were taken as poultry cuts that were processed during the visit or earlier before the visit with a total number of 183 samples that were drawn as follows:

One hundred and sixteen samples were taken as 4 samples from each of the 29 small scale places, which sell only freshly processed poultry carcases. Nineteen samples were taken from the 3 small scale places that had capabilities of chilling and freezing of poultry cuts. Twenty four samples were taken from the only large scale place included in the study. Twenty four samples were taken from the 2 places that were selling poultry cuts imported from Israel.

Samples were taken from places and put in a separate sterile plastic bag for every sample. Samples were put in an ice-box and immediately sent to the MOH, public Health Laboratory. Bacteriological analyses carried out in the Lab. Included presence of *Salmonella*, total plate count and count of both *Staphylococcus aureus* and *E. coli*.

4.10 Tools of Study

Three instruments were used for data collection

a. A structured face to face interview questionnaire was used for data collection.

b. Laboratory results were registered in a special form (Annex 9) before entering in Computer.

c. Bacteriological analysis carried out in the MOH Lab. was according to Bacteriological Analytical Manual 8th Edition of FDA, 1995.

4.11 Validity of the Instruments

The validation of the questionnaire was done by the researcher where distributing it to 10 different experts including researchers, managers, and specialised persons, they were

comments some notes which take care when writing the final questionnaire; the response rate of the experts was 80%, and finally minor change done according to implementations of pilot study. The methods of analysis are adopted by the USA, Food and Drug Administration so they were approved and there was no need for their validation.

4.12 Ethical Consideration

Several ethical considerations have been taken into account:

1- Consent form shall be signed by all persons included in the study (Annex 3) which includes the following:

- Participants have the right to voluntarily participate.
- Participants are previously informed about the study's objectives and data collected from them will be confidential and only for the purpose of the study.
- Participants have the right to withdraw from the study whenever they decide.
- Society's values, norms and cultures are respected during the whole study.

2- An agreement for using the Laboratory of the Ministry of Health should be concluded (Annex 1).

4.13 Construction of the Instrument

A structured face-to-face interview questionnaire was used as a tool for data collection (Annex 5) which focuses on:

Personal details for the Owner:

Personal, social and demographic data.

Health Status and practices of workers:

Personal hygiene practices.

Knowledge about *Salmonella*:

- If the person had ever contracted Salmonellosis.
- If the person has any knowledge about Salmonellosis in poultry.
- If the person has any knowledge about Salmonellosis in humans.

Slaughter Details:

- Address.
- Licensing validity.
- Source of water supply to the plant.
- Source of poultry intended for slaughter.
- Water used for scalding: its temperature, and its renewal.

Samples Details:

- Sample types.
- Source of the sample.
- Place of production and it is state.

Open Ended Question about the suggestions to improve production and marketing of poultry.

Attached the result of the Laboratory analysis.

The aim of the questionnaire is to determine the knowledge, practices and attitudes, of the workers in poultry slaughterhouses toward the application of Good Manufacturing Practices (GMP) concerning preparing the clean poultry for sale. Each owner of place was interviewed and accordingly 32 questionnaires were distributed to the head of workers or the keepership, researcher through face-to-face interviews completed the questionnaires with the workers in their workplaces and it takes about 20 minutes for each questionnaire.

4.14 MOH Public Health Laboratory

Established at 1994, work had started at 1996 in an extensive way-temporarily at Surani clinic. The lab. Moved to the new complex at Sabha clinic in the beginning of the year 2000, this complex was supported and founded by Italy. The lab. has full filled all the procedures and requiring according to ISO/IEC 17025. The lab activities involve to analyse:

1. Food microbiology and food chemistry.
2. Water microbiology and water chemistry.

4.15 Sample Collection

For laboratory examination, samples were collected from different locations during processing to determine the place in which *Salmonella* would probably contaminated during the processing. Fresh, chilled and frozen poultry meat samples were taken for microbial analysis *Salmonella* species and other bacteria from each slaughterhouse where the researcher was filling the questionnaire. The samples of chilled and frozen poultry meat were taken from the two main centres, which import such products from Israeli slaughterhouses; the sample unit consists of a minimum of 100 g. The 100g is used for preparation of the required standard 25g for microbial analysis, these samples taken, packed in clean sterile plastic sac, labelled with waterproof label, kept cold in ice box, attached with a special form as a request to the lab for analysis (Annex 7) and transported to the laboratory within one hour from collection. The samples sent for microbial analysis and to be examined as soon as they reached to the Laboratory, or it can be frozen at – 20°C for a period not exceeding 24 hours, and to be finally analyzed at the (MOH) Public Health Laboratory at Sabha Centre in Gaza City.

4.16 Preparation of the Sample

Twenty-five grams of each sample were homogenized at high speed in a stomacher with 225 ml peptone water (0.1%) for 2 minutes. Tenfold dilution was prepared under aseptic conditions from each sample using 0.1% peptone water as diluents. The diluted samples were used within 10 minutes after which they were discarded.

4.17 Equipment and Instruments on the MOH Public Health Laboratory in Gaza

1. Balance for Samples Preparation

Type: Sartorus

Modle:BP4100

Max capacity: 4100 g.

Readability: 0.1 g.

Reproducibility (Sd): $\leq \pm 0.1$ g.

2. Balance for Media Preparation

Type: Sartorus

Model: BP310S

Max: 310 g

Readability:0.001 g

Reproducibility (Sd): $\leq \pm 0.001$ g.

3. pH Meter

Type: HANNA

Model: HI 8520

Range pH: 0.00 to 14.00 pH

Temp. 0.0 to 100.0 ^{o}C

Resolution pH: 0.01 pH

Temp. 0.1 ^{o}C

Accuracy pH: ± 0.01 pH

Temp. ± 0.4 ^{o}C

4. Water Bath

Type: Techne

Model: TE- 8J

Operating temp range: 0 to + 85 ^{o}C

Temp Selection: Analogue

Temp Stability: ± 0.05 ^{o}C

Set point accuracy: ± 2% full scale range

Pump capacity:

Maximum Flow 5 L/ min.

Maximum Pressure 65 mbar

5. Autoclave

Type: KSG 40/60-2

Double- walled electrically heated

Operating pressure: 2.5 bar

Operating temp: 134 ^{o}C

6. Stomacher

Type: AES (Mix 1)

Motor speed: 240 rpm

Max bag capacity: 400 ml

Min bag capacity: 80 ml

Electronic timer adjustable from 10 sec. to 3 min

7. Incubator

Type: Memmert BE500

Working temp range: 5 ^{o}C to 70 ^{o}C

Temp set accuracy: 0.1 ^{o}C

Temp fluctuation: ± 0.1 ^{o}C

8. Utensils for Sample Handling and Processing such as:

Stainless Steel Knives of Various Lengths including one of 40 cm long.

Quebec Colony Counter, with Magnifying Lens

Sterile Plastic Polyethylene Bags (20-40 cm)

Sterile Plastic Polypropylene Cups (125 ml)

Sterile Plastic Polypropylene Syringes (1 ml and 10 ml)

Glassware

Plastic Ice box (30 l)

4.18 Bacteriological Analysis

The bacteriological media used throughout this study were prepared according to Difco (1985) and Oxoid (1995) manuals. The procedures followed for the isolation and counting

of different bacteriological parameters were in compliance with regulation of the Food and Drug Administration (FDA, 1995).

4.18.1 Media:

The following media were used throughout the study for the growth; differentiation and detection of bacteria see (Annex 5).

- Nutrient agar (Difco) for Heterotrophic Plate Count (H.P.C).
- Peptone water (Oxoid) for dilution.
- Bacto Violet red bile agar (Difco) *E. coli.*
- Bacto Baird Parker agar (Difco) for *Staphylococcus aureus*.
- Lactose broth (Difco) for *Salmonella* Pre enrichment.
- Selenite F- broth (Difco) for *Salmonella*, enrichment.
- S.S agar (Difco) for *Salmonella*.
- Bacto Bismuth sulfite agar (Difco) for *Salmonella*.
- Bacto Xylose Lysine Desoxycholate (XLD) agar (Difco) for *Salmonella*.
- Bacto Triple Sugar Iron (TSI) agar (Difco) for identification.
- Lysine Iron agar (Difco) for identification.
- Analytical profile index (API) 20E strips and API Staph (Bio Merieux), which can identify positive and negative results for *Salmonella* as shown in figure 2 below

A) API, Positive Result B) API, Negative Result

Figure 4.1: Positive and Negative Results of *Salmonella* According to Analytical Profile Index (API).

4.18.2 Bacteriological Procedures are Including the Following Testes (Annex 6):

- Aerobic total plate count (TPC).

- Isolation of *E. coli*.

- Isolation of *Staphylococcus aureus*.

- Isolation of *Salmonella*

4.19 Pilot Study

A pilot study was conducted before starting the actual data collection process, as pre-test for data collection instruments in order to test the suitability of the instruments and to detect the need of modifications to be done for the instruments and corrected to avoid biases and obstacles in implementation processes. The pilot study involved one main slaughter house, two small slaughter houses and one of the main supplier of chilled and frozen poultry meat. Nineteen samples were collected and analysed in MOH Public Health Lab, the pilot subjects were included in the study because the modification was light and all the samples were free of *Salmonella*.

4.20 Data Management

4.20.1 Data Entry:

- Designing a data entry model, through the statistical package for the social sciences program (SPSS) for the data collection instrument (questionnaire) in the computer.
- Questionnaires and the laboratory result analysis were overviewed.
- Data entry was done after the over viewing of the filled questionnaires.
- The data was cleaned through SPSS program, to ensure that all the data was entered correctly. This process was done through checking out a random number of the questionnaires and through conducting descriptive statistics and frequencies for all variables.

4.20.2 Data Analysis:

- Data analysis was done by the researcher with support from the supervisor, starting with the descriptive analysis, frequency tables were conducted for the study variables.
- Recording of certain variables.
- Cross tabulation for specific study variables.

Chi Square Statistical test was used to examine the relationship between variables. P value less than 0.05 considered statistically significant. Odds Ratio was used for measurement of risk and confidence interval was use for statistical significance testing.

4.21 Response Rate

The response rate in this study was 100%, all workplaces agreed to participate in the study. This indicates their interest in the topic to be researched.

4.22 Results of the Laboratory

The researcher collected the results of the laboratory analysis in the MOH Public Health lab form (Annex 10).

4.23 Limitation of the Study

1. The study is limited to Gaza city only as area of study.

2. Lack of funding.

3. There are no facilities in the lab to identifying the serotypes of *Salmonella* to compare with the similar studies.

Chapter 5

Results

CHAPTER 5

RESULTS

This chapter presents the results of the study and illustrates the descriptive analysis for the thirty-five work sites. These sites were authorized by Municipality of Gaza for slaughtering and selling poultry meat in Gaza City. The sites included thirty two small-scale sites for selling freshly slaughtered birds and one semi automated slaughterhouse for selling chilled and frozen poultry as well as two commercial centers selling chilled and frozen poultry meat. First, the researcher describe the study variables related to the workers and the sites of slaughtering; second, explore the bacteriological results; third, examine the relationship between *Salmonella* contamination and study variables and fourth summarize qualitative data about workers attitude to improve poultry production in Palestine.

5.1 Descriptive Analysis, Distribution of the Study Population

5.1.1 Socio Demographic Characteristics of the Study Population:

Table 5.1, summarizes workers' socio demographic characteristics, including age, gender, address, years of education, years of experience, and medical examinations.

Study population age ranged between 17 and 75 years old, these were divided into two groups: the first group included those workers of 35 years or less; it comprised 37.1% and the other group included those of older than 35 years old. All of the workers were males living in Gaza City.

Regarding years of education, illiterates were the lowest percent (5.7%), those with primary education were 17.2% yet, those with preparatory education were 31.4%, the highest percent was the secondary education (34.3%), and those with higher education were 11.4% of the study population. Table 5.1 shows two groups of education level: first group was equal or lees than nine years of education (54.3%), and the second group was more than nine years of education. Concerning experience of slaughtering and selling poultry, 51.4% of workers were working for more than ten years. Regarding medical examinations, the majority of workers (82.9%) medically tested for *Salmonella* in feaces and other tests.

Table 5.1: Distribution of the study population by socio demographic characteristics

Variable	No.	%	No.	%	Total	
					No.	**%**
Age group	≤ 35		> 35			
	13	37.1	22	62.9	**35**	**100**
Gender	Male		Female			
	35	100	0	0	**35**	**100**
Address	Gaza Governorate		Other Governorates			
	35	100	0	0	**35**	**100**
Years of education	≤ 9		> 9			
	19	54.3	16	45.7	**35**	**100**
Years of experience	≤ 10		> 10			
	17	48.6	18	51.4	**35**	**100**
Medical examinations	Yes		No			
	29	82.9	6	17.1	**35**	**100**

*** No. = Number**

5.1.2 Personal Hygiene:

The practical personal hygiene reflects workers behaviors, Table 5.2, Shows that all of workers cut theirs nails and most of them (97.1%) stay at home when they are infected with infectious disease, 94.3% dressing the wound with water proof cover, 97.1% don't wear gloves at work and the same percentage of workers (97.1%) does not wear jewelry while

working. In addition, 31.4% are wearing special clothes during work, the rest of them 68.6% return homes with the same working clothes. On the other hands 100% wash their hands with water and soap, after using water closet (WC), eating, and touching garbage, but 88.6% wash theirs hands at the end of working day, 11.4% after touching their hairs or work clothes, 13.6% after smoking, and 5.7% wash theirs hands at all mentioned situations.

Table 5.2: Distribution of the study population by personal hygiene of the workers

Variable	No.	%	No.	%	Total	
					No.	%
When gets infectious diseases	Stay working		Stay home			
	1	2.9	34	97.1	**35**	**100**
If wounded	Change place of work		Close dressing			
	2	5.7	33	94.3	**35**	**100**
Always nail cutting	Yes		N0			
	35	100	0	0	**35**	**100**
Wear special clothes at work	Yes		No			
	11	31.4	24	68.6	**35**	**100**
Return home wearing work clothes	Yes		No			
	24	68.6	11	31.4	**35**	**100**
Wearing gloves at work	Yes		No			
	1	2.9	34	97.1	**35**	**100**
Jewelry in hand during work	Yes		No			
	1	2.9	34	97.1	**35**	**100**
Hand washing with soap during work						
After use WC	Yes		No			
	35	100	0	0	**35**	**100**
After eating	Yes		No			
	35	100	0	0	**35**	**100**
After touching garbage	Yes		No			
	35	100	0	0	**35**	**100**
After smoking	Yes		No			
	3	13.6	19	86.4	**22***	**100**
After touching hair or clothes	Yes		No			
	4	11.4	31	88.6	**35**	**100**
Before work	Yes		No			
	22	62.9	13	37.1	**35**	**100**
At work end	Yes		No			
	31	88.6	4	11.4	**35**	**100**
All the above	Yes		No			
	2	5.7	33	94.3	**35**	**100**

*** Thirteen workers did not smoking (37.1% of study population)**

5.1.3 Workers Knowledge of *Salmonella*:

Table 5.3, shows workers knowledge about *Salmonella* where only 31.4% of workers heard about *Salmonella*, the majority of them (90.1%) heard through health education and the other heard through training. The study showed that 14.3% identified it as bacteria that cause human illness, while the rest (85.7%) did not know and 77.1% of study population did not know that poultry is considered a reservoir to *Salmonella* while 22.9% know that poultry intestines are considered the reservoir for Salmonella in the poultry carcass. About suffering from Salmonellosis, none answered that he suffered from *Salmonella*. The majority of the workers (94.3%) are sure that they did not suffer from *Salmonella* while 5.7% did not know if they suffered or not. 2.9% of their sons had get Salmonellosis, 80% don't know any one who had get Salmonellosis, and 17.1% did not know if any one of their family or public relation had got Salmonellosis.

Table 5.3: Distribution of the workers knowledge about *Salmonella* and suffering from salmonellosis

Variable	No.	%	No.	%	Total	
					No.	**%**
Heard about *Salmonella*	Yes		N0			
	11	31.4	24	68.6	**35**	**100**
If yes from where you known	Training		Health education			
	1	9.1	10	90.9	**11**	**100**
***Salmonella* is**	Bacteria		DK			
	5	14.3	30	85.7	**35**	**100**
Poultry as a source of *Salmonella*	Yes		No			
	8	22.9	27	77.1	**35**	**100**
Reservoir of *Salmonella* in poultry	Intestine		DK			
	8	22.9	27	77.1	**35**	**100**
Is *Salmonella* cause illness	Yes		No			
	8	22.9	27	77.1	**35**	**100**
Suffered from *Salmonella*	No		DK			
	33	94.3	2	5.7	**35**	**100**
Persons known suffered (*Salmonella*)	Yes		No			
	1	2.9	34	97.1	**35**	**100**
If yes whom	Son		Other			
	1	100	0	0	**1**	**100**

5.1.4 Localities of the Working Sites:

All sites of study population addressed in Gaza City, spread in eight areas Shajaia, Daraj, Tofah, Zaiton, Sabra, Rimal,Shati, and Shikh Radwan with percentages, 11.4%, 17%, 2.9%, 31.4%, 2.9%, 8.6%, 20%, 7.5% respectively; Zaiton is considered the highest area (31.4%) but Tofah and Sabra the lowest (2.9%).

5.1.5 Licensing and Inspection by the Official Organizations:

Workplaces of study population are authorized by Gaza Municipality, and there were different answers from the workers about official organizations who have responsibility of giving an agreement of license or make inspection in their sites. All of them say that the municipality gives agreement of license and makes inspection. Table 5.4, shows that there are 42.9% who have valid licenses for workplaces and the rest did not renew their license. Those workplaces sites were divided in two groups the first 15% did not renew theirs before the year 2000 and the second 85% have not renewed theirs since year 2000. Owners take agreements to license their workplace from MOH, MONE, and Civil Defense authority (82.9%, 5.7%, 20%) respectively. The workers receive medical examination in MOH laboratory. MOH also gives agreement to the municipality license for automatic slaughterhouse and main supplies of poultry meats that represent 8.6% of the total population study. Regarding the inspection, official organizations, MOH 14.3%, MONE 14.3%, and Civil Defense authority 8.6% had done this.

Table 5.4: Distribution of the study population by licensing of the workplaces

Variables	Frequency	Percentage
Workplace licenses still valid		
Yes	15	42.9
No	20	57.1
Total	**35**	**100**
Latest validity date for not valid license		
Before year 2000	3	15
At year 2000 and after	17	85
Total	**20**	**100**
Organizations give agreement for licensing		
Municipality	35	100
MOH	29	82.9
MONE	2	5.7
Civil Defense	7	20
Organizations make inspection		
Municipality	35	100
MOH	5	14.3
MONE	5	14.3
Civil Defense	3	8.6

5.1.6 Environment at Working Sites:

Table 5.5 and 5.6, summarized the environment situation at workplace; it shows that all of workplaces connected with municipal sewage network, and no one runs liquid waste out side the workplace and solid waste is daily transferred from workplaces and municipal garbage boxes to the landfill.

The environment situations at workplaces are good with percentage 11.4%, 51.4% are accepting, and 37.1% are rejecting. The rejection was because 61.5% cages or the poultry were outside the workplace, and 38.5% also rejected because the place was not suitable because of garbage surrounding workplace and not transferred to the municipality garbage box. Regarding suitability of workplace building, the space of 62.9% is enough, the space of 37.1% is not enough for working need; there were different types of ceiling concrete,

asbestos, and metals. The majority is concrete (74.3%). About the floor of workplace, it was 91.4% smooth and 8.6% was unacceptable because of wholes or creaks.

Airing depend on natural and artificial way; 97.1% depend on both natural and artificial ways, 2.9% depend on artificial way only, and non depend on natural way only, airing quality 11.4% good and 88.6% accepted.

Table 5.5: Distribution of the environment situation at workplaces

Variable	No.	%	No.	%	Total	
					No.	%
Environment outdoor workplace	Good		Not good			
	4	11.4	31	88.6	**35**	**100**
Reasons of not accepted	Chicken cage at street		Garbage not transferred			
	8	61.5	5	38.5	**13**	**100**
Square place of work	Enough		Not enough			
	22	62.9	13	37.1	**35**	**100**
Type of ceiling	Concrete		Asbestos or Metal			
	26	74.3	9	25.7	**35**	**100**
Floor of workplace	Good		There is broken floor			
	32	91.4	3	8.6	**35**	**100**
Type of ventilation	Artificial		Natural &artificial			
	1	2.9	34	97.1	**35**	**100**
Ventilation quality	Good		Accept			
	4	11.4	31	88.6	**35**	**100**
Type of light	Natural &artificial		Other			
	35	100	0	0	**35**	**100**
Light quality	Good		Accept			
	9	25.7	26	74.3	**35**	**100**

Regarding water supply, all workplaces had municipal water supply, except for 2.9% that had received water from municipal water and a private well, 88.6% had enough water, all of workplaces are provided with firmly closed water tanks, majority (91.4%) of workplaces had plastic tanks and the rest had concrete or metal tanks. The lengths of municipal garbage box from workplaces are equally or less than 350 meters are 48.6%, and the rest are longer than 350 meters. Solid waste transferred by workers in the workplaces with

percent 42.9% and the rest transferred by municipal workers. Regarding removing blood from workplace, 45.5% of study population removes blood through sewage net and the rest remove it through solid waste garbage boxes.

Table 5.6: Distribution of the water supply and garbage removal at workplaces

Variable	No.	%	No.	%	Total	
					No.	%
Source of water	Municipality		Special well			
	35	100	1	2.9		
Enough supply water	Yes		No			
	31	88.6	4	11.4	**35**	**100**
Storage water at place	Yes		No			
	35	100	0	0	**35**	**100**
Type of water storage material	Plastic		Concrete or Metal			
	32	91.4	5	14.3		
Water storage good closed	Yes		No			
	35	100	0	0	**35**	**100**
Connected with sewage net	Yes		No			
	35	100	0	0	**35**	**100**
Throw sewage out of the net	Yes		No			
	0	0	35	100	**35**	**100**
Nearest municipal garbage box	≤ 350		> 350			
	17	48.6	18	51.4	**35**	**100**
Solid waste removed to municipal garbage box	Place workers		Municipality labors			
	9	25.7	26	74.3	**35**	**100**
Workplace removing blood through	Sewage net		Municipal garbage			
	15	45.5	18	54.5	**33**	**100**

5.1.7 Processing at Place of Work:

All workplaces, which have slaughtering or handling poultry meat, never use chlorine as disinfectant in carcass cleaning water nor use radiation method, but all of them used plastic packaging material. No freezers and refrigerators include recording temperature instrument to show quality through a month. Table 5.7, 5.8 and 5.9 shows that 65.7% used pests control methods; most of them (82.6%) use chemicals and the rest are equal in using ultra violet (UV) and mechanical methods (8.7%). 97.1% use detergents, but users of hot water,

paste soap, liquid soap and disinfectant like chlorine are (79.4%, 57.1%, 54.3%, 31.4%) respectively. All slaughterhouses cleaning their equipments and machines as mentioned above but 2.9% do not clean because they are selling prepackaged chilled and frozen poultry products without opening the packages. The one who makes clean for equipments and machines he does cleaning once, twice or more (8.8%, 41.2%, 50%) respectively. All small slaughterhouses get rid of feathers by using manual mechanical method with hot water, but the automatic slaughterhouse represent 3% of the slaughtering house place feathering automatically with tap water. All small slaughterhouses control scalding water temperature by personal sensory and change it once, twice and more than two (6.3%, 53.1%, and 40.6%) respectively.

About cleaning and washing carcass after feathering and removing intestines, they use running water 51.5% and the rest use water in sink several times. There are only 57.1% who sell fresh poultry meat, 5.7% are selling only chilled and frozen poultry and the rest are selling fresh, chilled and frozen poultry. Sellers of chilled poultry meat, 64.3%, have refrigerators capacity as 1-10 m^3 and the rest of them have refrigerators with capacity of more than $10m^3$. Twenty percent are selling frozen poultry, 11.4% have freezer capacity of 1-3 m^3 and 8.6% have freezer with capacity of more than 100 m^3 and with temperature clock.

The majority of slaughterhouse (97%) may leave live poultry for more than a day at the workplace, and the rest of them are slaughtering poultry on the same day and then separate slaughtered cleaned (feathered and removed intestine) poultry away from slaughtering and cleaning place. Only 5.7% of study population freezing poultry at their workplaces, 50% of them has freezer with capacity 34 m^3 its temperature (-25°C) and the rest have small freezer (1 m^3) with temperature (-18°C) which is considered unsuitable for commercial freezing.

Table 5.7: Distribution of the pests control and cleaning at workplaces

Variable	No.	%	No.	%	Total	
					No.	**%**
Workplace protected against pests	Yes		No			
	6	17.1	29	82.9	**35**	**100**
Use methods for controlling pests	Yes		No			
	23	65.7	12	34.3	**35**	**100**
Type of methods used to control pests	Chemical		Mechanical & UV.			
	19	82.6	4	17.4	**23**	**100**
Storage of Chemicals in special place	Yes		No			
	19	100	0	0	**19**	**100**
Use detergents in cleaning equipments	Yes		No			
	34	97.1	1	2.9	**35**	**100**
Use disinfectant like chlorine in cleaning	Yes		No			
	11	31.4	24	68.6	**35**	**100**
Use paste soap	Yes		No			
	20	57.1	15	42.9	**35**	**100**
Use liquid soap	Yes		No			
	19	54.3	16	45.7	**35**	**100**
Clean machines and equipments	Yes		No			
	34	97.1	1	2.9	**33**	**100**
Use hot water in cleaning	Yes		No			
	27	79.4	8	20.6	**19**	**100**
Repeat cleaning daily machines & equipment	≤ 2		>2			
	17	50	17	50	**34**	**100**
Cleaning the carcass by water	water used once		Rebate use same water			
	17	51.5	16	48.5	**33**	**100**
Using chlorine in carcass water cleaning	Yes		No			
	0	0	33	100	**33**	**100**
Use radiation in sterilization	Yes		No			
	0	0	35	100	**35**	**100**

Table 5.8: Distribution of the study population by feathering at workplaces

Variable	No.	%	No.	%	Total	
					No.	%
Poultry feathering	Machine manual		Automatically			
	32	97	1	3	**33**	**100**
Water used in defeathering	Hot water		Tap water			
	32	97	1	3	**33**	**100**
Control hot water temperature	Yes		No			
	32	97	1	3	**33**	**100**
Change feathers water daily	≤ 2		>2			
	19	59.4	13	40.6	**32**	**100**

Table 5.9: Distribution of the study population by handling chilled or frozen poultry at workplaces and using packaging material

Variable	No.	%	No.	%	Total	
					No.	%
Sale chilled or frozen poultry	Yes		No			
	15	42.9	20	57.1	**35**	**100**
If selling area separate from poultry preparation	Yes		No			
	2	15.4	11	84.6	**13**	**100**
Capacity of the chilling refrigerator (meters^3)	≤ 10		> 10			
	9	60	6	40	**15**	**100**
Freezing poultry in the Workplace	Yes		No			
	2	5.7	33	94.3	**35**	**100**
If yes what it is capacity (m^3)	1 m^3		34 m^3			
	1	50	1	50	**2**	**100**
Temperature of freezing(°C)	-25		-18			
	1	50	1	50	**35**	**100**
Have refrigerator for storage frozen poultry	Yes		No			
	7	20	28	80	**35**	**100**
Capacity of freezing product refrigerator (m^3)	1-3		≥ 100			
	4	57.1	3	42.9	**7**	**100**
Temperature of storage frozen poultry	-18°C		Other			
	7	100	0	0	**7**	**100**
Refrigerator has a watch to identify the temperature	Yes		No			
	3	42.9	4	57.1	**7**	**100**
Recording temperature of the refrigerator	Yes		No			
	0	0	7	100	**7**	**100**

5.1.8 Source of Poultry and Poultry Meat:

The major source of poultry and poultry meat (94.3%) as shown in Table 5.10 is the Gaza Strip Governorates, which include all small slaughterhouses that comprised 91.4% of study population source of their live poultry are from Gaza Strip Governorates alone. While 8.6% get their goods from Israeli side that include 2.9% their sources from Gaza Strip Governorates and Israeli side who import chilled turkey meat and live turkey.

Only 3% of study population segregate live poultry from processing area or and final products; these slaughter their poultry once they receiving it and not holding it in workplace.

Table 5.10: Distribution of the source of goods and receiving places at workplaces

Variable	No.	%	No.	%	Total	
					No.	%
Gaza strip Governorates	Yes		No			
	33	94.3	2	5.7	**35**	**100**
Others (Israel)	Yes		No			
	3	8.6	32	91.4	**35**	**100**
Live poultry segregated from processing final product	Yes		No			
	1	3	32	97	**33**	**100**
Longer holding time of live poultry in workplace	One day		More than one day			
	1	3	32	97	**33**	**100**

5.1.9 Poultry Meat Samples:

The state of 183 poultry samples, which were collected from Gaza markets and tested for microbiological analysis in MOH Public Health Laboratory, is as follows: 64.5% chicken meat, 12.6% turkey meat, 18.6% edible chicken offal, and 4.4% edible turkey offal. The origin of live poultry, is considered the source of the analyzed samples, which were taken from North Gaza, Gaza, Mid-zone, South Gaza and Israel (16%, 24.6%, 4.4%, 47%, 15.3%) respectively. Table 5.11, shows that major sample is chicken (83%), source site of

the samples (86.88%) was local slaughtering sites, and the rest from Israel. The fresh state samples were higher (68.3%) than chilled and frozen samples (31.7%).

Table 5.11: Distribution of the description of poultry samples which were collected from workplaces

Variable	No.	%	No.	%	Total	
					No.	%
Sample type	Chicken		Turkey			
	152	83	31	17	**183**	**100**
Sample origin (live, chilled & frozen poultry)	Gaza Governorates		Other (Israel)			
	155	84.7	28	15.3	**183**	**100**
Producer site of the sample	Local workplace		Israel			
	159	86.88	24	13.11	**183**	**100**
Sample state	Fresh		Chilled & frozen			
	125	68.3	58	31.7	**183**	**100**

5.2 The Microbiological Analysis Results

Palestinian Standard, not allow presence of *Salmonella* at fresh and chilled poultry, total plate count (TPC) must not increase more than 5×10^5 /1 gram, but frozen poultry TPC must not increase more than 2.5×10^5/1 gram. Table 5.12, shows that not compliance poultry samples which have high level of TPC than acceptable level has the lowest percent (5.5%) comparing with not compliance in other bacteriological analysis according PS. Chilled samples are higher contaminating with TPC (21.9%) than fresh and frozen samples. The overall frequency of *Salmonella* contamination in poultry samples is 16.4%; fresh samples are higher contaminated (19.2%) than chilled samples while frozen samples are free of *Salmonella*.

Staphylococcus aureus and *Escherichia coli* not include in the Palestinian Standards of fresh, chilled poultry, and frozen poultry standards, but include in Palestinian Standards of fresh chilled meat, the maximum acceptable criteria to both *Staphylococcus aureus* and

Escherichia coli are 50/1 gram, and 100/1 gram respectively. When we applying the same criteria on our samples it was found out as Table 5.12, shows that (12%,) of poultry samples rejected because of the over count of *Staphylococcus aureus* and chilled samples are higher contaminated (25%) than fresh and frozen samples. Poultry samples rejected because of the over count of *Escherichia coli* is (25.1%), chilled samples are higher contaminated (34.4%) than fresh samples while all frozen samples are whiten acceptable level. In addition, there are 19.13% of study population samples fail in *Salmonella* and TPC analysis, and 40.44% of samples consider fail at the fourth analysis.

Table 5.12: Distribution of the bacteriological analysis of the poultry samples according state of sample*

Sample State	Bacteriological Result			
	Accept		Reject	
	No.	%	No.	%
Total plate count				
Fresh	122	97.6	3	2.4
Chilled	25	78.1	7	21.9
Frozen	25	96.2	1	3.8
Total	**173**	**94.5**	**10**	**5.5**
Salmonella				
Fresh	101	80.8	24	19.2
Chilled	26	81.3	6	18.8
Frozen	26	100	0	0
Total	**153**	**83.6**	**30**	**16.4**
Staphylococcus aureus				
Fresh	111	88.8	14	11.2
Chilled	24	75	8	25
Frozen	26	100	0	0
Total	**161**	**88**	**22**	**12**
Escherichia coli				
Fresh	90	72	35	28
Chilled	21	65.6	11	34.4
Frozen	26	100	0	0
Total	**137**	**85.8**	**46**	**25.1**
Total of analysis	**109**	**59.56**	**74**	**40.44**

***Classification is accepting criteria based on Palestinian standard**

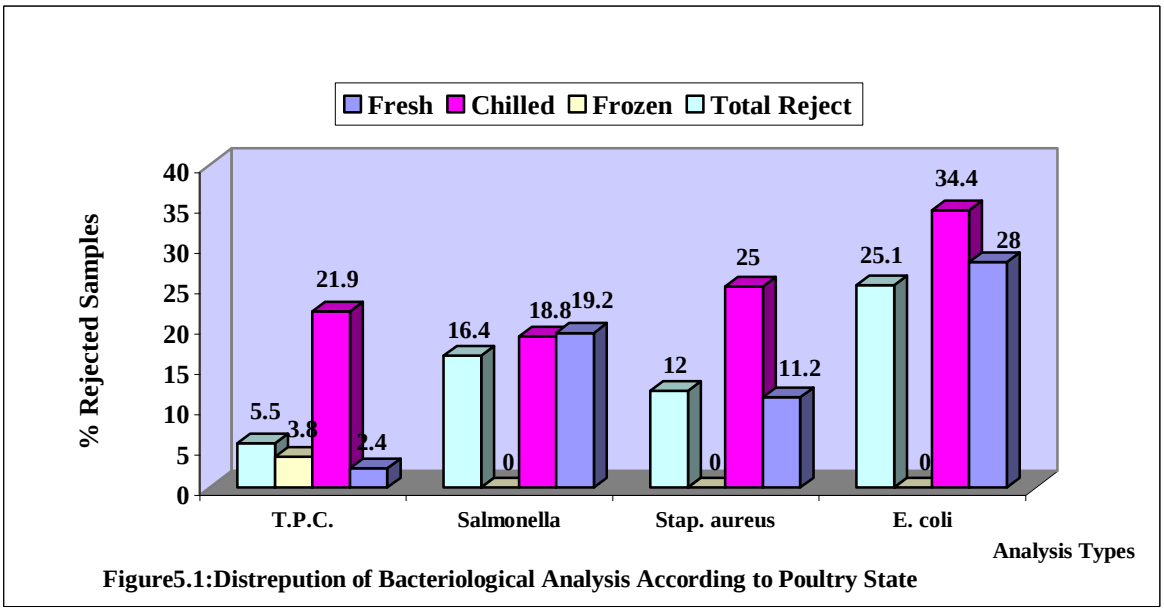


Figure5.1:Distreputionof Bacteriological Analysis According to Poultry State

In general, bacteriological results for samples of fresh, chilled and frozen poultry shows that chilled poultry were the most contaminated, while frozen poultry were the least contaminated and were free of *Salmonella*. This situation can be due to the fact frozen poultry were packaged and packaging prevents cross-contamination and freezing temperature prevents the growth of bacteria. In the case of chilled poultry, high contamination can be referred to being not packaged that leads to more cross-contamination from handlers and other poultry carcasses in addition to inadequate storage temperature that allows growth of bacteria. In case of fresh poultry, contamination referred to inadequate hygienic practices namely cleaning and washing.

These findings comply with the findings of a study where 75% of chicken carcasses purchased in supermarkets and 25% of those purchased from poultry shops were contaminated with *Salmonella* (Capit`a, et al., 2003). But it does not comply with another study that found no significant difference between fresh retail and frozen chicken carcasses contaminated with *Salmonella* (Meldrum, et al., 2004). In addition, Food Standards

Agency in UK (2003) showed fresh chicken to have lower *Salmonella* contamination (4%), compared to frozen chicken (10.4%). The results in present study were different from that in UK because of improper handling, especially for fresh and chilled poultry that increase contamination.

5.3 Inferential Analysis

The following results explore the relationships between knowledge, practices and source of poultry samples with presence of *Salmonella* in poultry meat.

5.3.1 Distribution Personal Characteristics and Knowledge of Workers by Presence of *Salmonella* in Poultry:

Table 5.13 shows the distribution of personal characteristics and presence of *Salmonella* in poultry. It can be noticed that workers aged less than 30 years old were related to more positive samples (26.4%) than older workers (15.1%). The difference between these two age groups did not reach a statistical significant level (P = 0.09).

Workers who had education level equal or less than 9 years had less positive samples for *Salmonella* (18.6%) than other workers (19.4%). However that difference was not statistically significant (P = 0.9). Workers who have experience in field slaughtering or selling poultry period equal or less than 10 years have less presence of *Salmonella* in their poultry samples (18.4%) than other workers (19.1%), the difference between two groups did not reach a statistical significant (P= 0.91). Workers who wear special clothes during working their poultry samples have less contamination with *Salmonella* (12.1%) than others (23.7%) but the variations between the two groups did not reach a statistical significant level (P= 0.07). Workers who apply hand washing rule with soap during work according to requirement have less contamination with *Salmonella* (8.3%) than others

(20.7%) but the variations between the two groups did not reach a statistical significant level (P= 0.15). Workers did not make medical handler examination on their poultry samples have more contamination with *Salmonella* (28%) than other workers (17.2%) the difference between two groups did not reach a statistical significant difference.

Table 5.13: Distribution personal characteristics (age, educational level, experience, wearing special clothes, hand washing, and medical examination) of workers by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R. *		P. value
	Positive		Negative				
	No*.	%	No.	%	Value	CI*	
Workers age							
≤ 35	14	26.4	39	73.6	2.02	0.9 – 4.54	0.09
> 35	16	15.1	90	84.9			
Education level							
≤ 9	18	18.6	79	81.4	0.95	0.42 – 2.14	0.90
> 9	12	19.4	50	80.6			
Experience period							
≤ 10	9	18.4	40	81.6	0.95	0.4 – 2.3	0.91
> 10	21	19.1	89	80.9			
Wearing special clothes							
Yes	8	12.1	58	87.9	0.45	0.19 – 1.07	0.07
No	22	23.7	71	76.3			
Hand washing with soap during work according rule requirement							
Yes	2	8.3	22	91.7	0.35	0.08 - 1.57	0.15
No	28	20.7	107	79.3			
Medical examination							
Yes	23	17.2	111	82.8	0.53	0.20 – 1.42	0.20
No	7	28	18	72			

*** O.R = Odds ratio, *No. = Number,*CI = Confidence interval**

Workers aged more than 35 years had less *Salmonella* present in their poultry(15.1%) compared to workers less than 35 years old (26.4 %). This can be due to older workers had more experience and had low education that facilitated their obedience to hygienic regulations. Some authors mentioned that more caring workers reduced contamination (Ishii et al., 1989; Cheng and Beuchat, 1995; Federighi et al., 1995 and Capita et al., 2000)

and they found that increasing accuracy and care in removal of intestine led to reduced contamination.

Handlers obeying personal hygiene practices, such as wearing special clothes, washing hands with soap during work and passing mandatory medical examinations had less poultry contaminated with *Salmonella* than others who did not comply with such regulations.

Unfortunately, there are no available studies about effects of handlers' personal characteristics such as age, education level, and experience period as risk factors of prevalence of *Salmonella* in poultry carcasses.

Regarding workers knowledge about *Salmonella*, Table 5.14 reveals workers knowledge about *Salmonella*. Workers who heard about *Salmonella* the presence of *Salmonella* in their poultry samples (8%) less than other workers(22%), the difference between two groups did not reach a statistical significant (P= 0.24). Workers who know that *Salmonella* cause human illness have less *Salmonella* presence in their poultry samples (14.3%) than other group that do not know (20.9%), but the difference between two groups is not statistically significant (P= 0.32). Workers who know that poultry could be source of *Salmonella* has less *Salmonella* presence in their poultry samples (14.3%) than other group that do not know (18.9%), but the difference between two groups did not reach a statistical significant difference

Table 5.14: Distribution of workers knowledge about *Salmonella* by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
Workers hearing *Salmonella*							
Yes	8	14	49	86	0.59	0.25– 1.44	0.24
No	22	21.6	80	78.4			
***Salmonella* cause illness**							
Yes	7	14.3	42	85.7	0.63	0.25 - 1.59	0.32
DK	23	20.9	87	79.1			
Poultry could be source of *Salmonella*							
Yes	7	14.3	42	85.7	0.63	0.25 - 1.59	0.32
DK	30	18.9	129	81.1			

The study showed that workers' knowledge about *Salmonella* has a positive effect on reduction of *Salmonella* contamination in poultry carcasses during handling and preparation, because knowing hazard makes people becoming more aware.

5.3.2 Distribution Sites, License, Inspection, Environment and Pest Control of Workplace by Presence of *Salmonella* in Poultry:

Workplace sites of study population in Gaza city can be divided into three areas: first Shajaia, second Gaza[1] (include Zaiton, Daraj, Tofah, Sabra and Remal) and third Shati and Sh.Rodwan, the lowest samples contaminated with *Salmonella* is Gaza[1] area (11.7%), and the highest samples contaminated with *Salmonella* are Shati & Sh. Rodwan area (36.1%). As shown in Table 5.15. When we take Gaza[1] as reference area for the other two areas we found that samples of Shajaia area has high presence of *Salmonella* (33.3%) than Gaza[1] (11.7%), the difference between Gaza[1] and Shajaia is not statistically significant (P= 0.06). However, Shati & Sh.Rodwan samples has high presence of *Salmonella* (36.1%) than Gaza[1] (11.7%), the difference between the two areas are statistically significant (P= 0.001).

Workplace with valid license has high presence of *Salmonella* (24.1%) than not valid license (17%) but the difference between the two places did not reach a statistical significant difference. Although inspection in place of work by MOH or MONE reduce presence of *Salmonella* (13.5%)than places which were not inspected of by MOH or MONE (20.5%) but the difference between the two groups is not statistically significant (P=0.34). Workplaces with good outdoor environmental status have less contamination poultry with *Salmonella* (5.6%) than other worksites (22.8%); the difference between the two groups is statistically significant (P = 0.02). Place of work closed carefully against pests had more contaminated poultry samples with *Salmonella* (37.5%) than other places of work (16.8%), the difference between the two groups is statistically significant (P=0.045). Workplaces which applies pests control by using chemicals have more contaminated poultry sample with *Salmonella* (27.7%) than other places which did not make pest control (11.4%), the difference between the two groups is statistically significant (P=0.034). Place of work which apply pests control, by using ultra violet (UV) and mechanical methods have less contaminated poultry sample with *Salmonella* (6.25%) than other places which did not make pest control (11.4%) the difference between the two groups is not statistically significant (P=0.45).

Table 5.15: Distribution sites, license, inspection, environment and pest control of workplace by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
Workplace address							
Shajaia	4	33.3	8	66.7	3.77	0.81– 16.77	0.06
Gaza[1]	13	11.7	98	88.3	1		
Shati& Sh.Rodwan	13	36.1	23	63.9	4.26	1.6 – 11.4	0.001 *
License validity							
Yes	13	24.1	41	75.9	1.64	0.73 – 3.7	0.23
No	17	17	88	83.8			
Inspection in place of work by MOH or MONE							
Yes	5	13.5	32	86.5	0.61	0.58 – 4.67	0.34
No	25	20.5	97	79.5			
Environment outdoor							
Good	2	5.6	34	94.4	0.20	0.05 – 0.88	0.02 *
Not good	28	22.8	95	77.2			
Place of work closed carefully against pests							
Yes	6	37.5	10	62.5	2.98	0. 98 – 8.97	0.045 *
No	24	16.8	119	83.2			
Pest control methods							
Chemical	23	27.7	60	72.3	2.99	0.97 – 9.85	0.034 *
No pest control	5	11.4	39	88.6	1		
UV& Mechanical	2	6.25	30	93.75	0.52	0.06 – 3.36	0.45

Gaza[1] = include Zaiton, Daraj, Tofah, Sabra and Remal

*** Statistically significant**

Geographically, Shati Camp and Sh. Radwan areas had the highest contamination with *Salmonella*, next was Shajaia area, while Gaza[1] area (Zaiton, Daraj, Tofah, Sabra and Remal) had the lowest contaminated poultry samples. That can be referred to Gaza[1] area has a good environmental conditions in out door workplace, workers had more awareness and experience compared with those in Shati camp, Sh.Rodwan and Shajaia areas. The geographical difference, between areas in Gaza City, was a statistically significant. That finding agrees with the survey of Food Standards Agency in UK (2003), which revealed

significant differences between the four countries (Wales, England, Northern Ireland and Scotland) in the UK about frequency of *Salmonella* contamination in retail chicken.

Paradoxically, licensed workplaces had more poultry contaminated with *Salmonella* than places without valid license. This may be attributed to the notice that licensed places have bad outdoor environment, inadequate cleaning and disinfecting of equipment, workers were less experienced and in the same place slaughtering and processing rabbits, ducks, geese, pigeon and quail. Workplaces, including automatic slaughterhouse and main suppliers, that were under inspection by official organizations as MOH and MONE had less contamination compared to other places (small slaughter house). This can be due to large workplace and periodical inspection, increase handlers awareness.

Places applying pest control by Ultraviolet light had least contamination in poultry compared to places employing chemicals and places not applying pest control at all. This can be referred to miss-using of chemicals, and place contamination by inadequate cleaning. In addition, Ultraviolet has disinfecting effects on equipment surfaces.

5.3.3 Distribution Management Principals of Workplace by Presence of *Salmonella* in Poultry:

Table 5.16, show that workplaces which have poultry inspection by official organization have less contamination poultry with *Salmonella* (8.3%) than other worksites(20.7%); the difference between the two groups is not statistically significant (P = 0.15). In addition, we have the same result; fewer samples contaminated with *Salmonella* in case of segregation live birds from feathering area and final product, feathering automatically with tape water, have a source of hot water, and did not retain live poultry to another day in workplace because these properties found only in the automatic slaughtering site. Workplaces which

isolate diseased poultry from non-diseased poultry have less contamination poultry with *Salmonella* (18.7%) than other work sites (25%); the difference between the two groups is not statistically significant (P = 0.75). Workplaces which slaughtering chicken and turkey poultry have less contamination poultry with *Salmonella* (17.5%) than other work sites which slaughtering chicken, turkey and other poultry (24.2%), the difference between the two groups is not statistically significant (P = 0.38).

Workplaces which remove solid waste by them self have less contamination poultry with *Salmonella* (15.1%) than other work sites which depend on municipal workers (22.1%) the difference between the two groups is not statistically significant (P = 0.26). Workplaces which remove blood of slaughtering poultry to sewage net have more contamination poultry with *Salmonella* (20.9%) than other worksites which remove blood of slaughtering poultry to municipal garbage boxes (17.4%), the difference between the two groups is not statistically significant (P = 0.58). Workplaces, which Selling frozen or/and chilled poultry, have less contamination poultry with *Salmonella* (17.7%) than other worksites (20%); the difference between the two groups did not reach a statistical significant difference. In addition, workplaces, which freeze poultry have less contaminated poultry with *Salmonella* (7.1%) than other places (21.4%), the difference between the two groups is not statistically significant (P=0.08).

Table 5.16: Distribution management principals of workplace by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
Poultry inspected by official organization							
Yes	2	8.3	22	91.7	0.35	0.08 – 1.57	0.15
No	28	20.7	107	79.3			
Isolated disease poultry							
Yes	29	18.7	126	81.3	0.69	0.07 – 6.88	0.75
No	1	25	3	75			
Poultry types slaughtering							
Chicken & turkey	22	17.5	104	82.5	0.66	0.26 – 1.66	0.38
Chicken, turkey &other	8	24.2	25	75.8			
Responsibility of removing solid waste							
Owners	11	15.1	62	84.9	0.63	0.28 – 1.42	0.26
Municipality worker	19	22.1	67	77.9			
Removal blood							
Sewage net	14	20.9	53	79.1	1.26	0.57 – 2.79	0.58
Municipal garbage box	16	17.4	76	82.6			
Selling frozen or/and chilled poultry							
Yes	14	17.7	65	82.3	0.86	0.39 – 1.91	0.71
No	16	20	64	80			
Freezing poultry							
Yes	2	7.1	26	92.9	0.28	0.06 – 1.27	0.08
No	28	21.4	103	78.6			

Results of workplaces under regular supervision of MOH and MONE inspectors had less contamination than other places. These places segregate live birds from processing area and from final product, remove feather with tape water automatically and have a source of hot water. These places also have good facilities, their products are under control and they comply with Palestinian Standards.

Slaughterhouse that apply official recommendations as isolating ill birds away, and processing only chicken and turkey, had less contamination. This may be due to preventing cross-contamination from ill poultry and others domestic poultry (duck, geese, pigeon, rabbits and quail) that may be infected with *Salmonella*.

Workplaces that transport solid wastes by themselves had less contaminated poultry than other places that depend on municipal workers. This can be referred to, more appropriate and timely transport by them.

Paradoxically, results showed that removing blood of slaughtered poultry to sewage net increased contamination than removing it to municipal garbage boxes. It can be assumed that removing blood to sewage net leads to closing by blood clots, which in turn leads expected flooding of sewage into workplace. This reflects workers' personal carelessness.

Workplaces that freeze poultry had less contamination than other places. This can be caused by, effects of freezing temperatures and preparation of poultry such as cleaning and packaging in suppressing bacterial growth and consequently reducing the contamination.

5.3.4 Distribution Workplace, Water Supply, and Cleaning with Detergent and Disinfectant by Presence of *Salmonella* in Poultry:

Table 5.17, shows that workplaces which have enough water supply have less contamination poultry with *Salmonella* (18.2%) than other work sites which have scarcity of water (25%), the difference between the two groups is not statistically significant (P = 0.51).

Workplaces which use paste detergent have more contaminated poultry samples with *Salmonella* (26.9%) than other places which did not used paste detergent (11.1%), the difference between the two groups is statistically significant (P=0.01). While workplaces which use liquid detergent have less contaminated poultry samples with *Salmonella* (13.8%) than other places which did not used liquid detergent(26.2%), the difference between the two groups is statistically significant (P=0.05). Workplaces which use chlorine disinfectant in cleaning equipment have less contaminated poultry samples with

Salmonella (12.9%) than other places (23.6%), the difference between the two groups is not statistically significant (P=0.09).

Workplaces, which clean equipment and machine more than twice daily have more contaminated poultry samples with *Salmonella* (21.3%) than other places which clean equipment and machine twice or less daily(16.7%). The difference between the two groups, did not reach statistical significant difference. Also workplaces which changing feathering water more than twice daily have more contaminated poultry samples with *Salmonella* (22%) than other places which changing feathering water twice or less daily(19.7%), the difference between the two groups is not statistically significant (P=0.74).

Table 5.17: Distribution workplace water supply, and cleaning with detergent and disinfectant by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
Enough water supply							
Yes	26	18.2	117	81.8	0.67	0.2 – 2.23	0.51
No	4	25	12	75			
Paste detergent							
Yes	21	26.9	57	73.1	2.95	1.25 – 6.93	0.01 *
No	9	11.1	72	88.9			
Liquid detergent							
Yes	13	13.8	81	86.2	0.45	0.20 – 1.01	0.05 *
No	17	26.2	48	73.8			
Using chlorine disinfectant in cleaning equipment							
Yes	9	12.9	61	87.1	0.48	0.20 – 1.12	0.09
No	21	23.6	68	76.4			
Repeating cleaning equipment & machine							
≤ 2	14	16.7	70	83.3	0.74	0.33 – 1.64	0.45
>2	16	21.3	59	78.7			
Changing feathering water							
≤ 2	15	19.7	61	80.3	0.87	0.38 – 2.01	0.74
> 2	13	22	46	78			

*** Statistically significant**

Results of workplaces provided with enough water showed less contamination than other places. It must be remembered that water is considered as most important material for cleansing of equipment and the leads to reduced contamination.

Employing liquid detergent reduced contamination. Liquid detergents easily spread and make efficient solutions for removing dirt, but paste detergents increase contamination, because diffusion in water is not adequate and lead to spreading of dirt and increase contamination. In addition, it lost in wastewater.

Cleansing of equipment and feathering machines more than twice-daily increased contamination. This can be referred to inadequate cleansing and not employing disinfectant materials.

5.3.5 Distribution Poultry Sample Characteristics by Presence of *Salmonella* in Poultry:

Table 5.18, shows that poultry samples theirs origin Gaza and north Gaza Governorate have higher contamination with *Salmonella* (21.3%) than others from out of Gaza and north Gaza Governorate (13.9%), the difference between the two groups did not reach statistical significant difference. Chicken samples have higher contamination with *Salmonella* (19.1%) than turkey (3.2%), the variations between the two types is statistical significant (P=0.03). Fresh poultry have light higher contamination with *Salmonella* (19.2%) than chilled poultry (18.8%), the difference between the two groups did not reach statistical significant difference, while all frozen poultry samples are free of *Salmonella*.

Table 5.18: Distribution sample characteristics by presence of *Salmonella* in poultry

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
Sample origin							
Gaza & North	13	21.3	48	78.7	1.67	0.73 – 3.72	0.20
Out of above	17	13.9	105	86.1			
Type of sample							
Chicken	29	19.1	123	80.9	7.1	0.93 – 54	0.03 *
Turkey	1	3.2	30	96.8			
Sample state							
Fresh	24	19.2	101	80.8	1.03	0.35 – 3.15	0.95
Chilled	6	18.8	26	81.3			

*** Statistically significant**

Poultry raised in Gaza City and North Gaza Governorate had higher contamination than poultry raised elsewhere. This assumed to be due to inefficient disinfection of farms between stockings in Gaza City and North Gaza compared to that practiced elsewhere. Turkey samples had less contamination than chicken. This can be referred to breeding turkeys costs high, need longer period of time, take more care and requires special skill and more drug than chicken. Those reasons may reduce prevalence of *Salmonella* in live turkey. In addition, most of turkeys slaughtered in the automatic slaughterhouse, which applies hygienic conditions and is under official control. This finding is in agreement with Safwat et al., 1985 who found more *Salmonella* in chicken meat (9.05%) than in turkey meat (3.4%), and also with Khosrof Ben Jaafar, et al., 2002 who found 3.6% of chicken meat, but only 1.7% of turkey meat to be contaminated with *Salmonella*. Fresh poultry had higher contamination than chilled poultry, which can be assumed to be due to fresh poultry received less cleansing.

5.3.6 Distribution of Poultry Samples Produced by Automatic Slaughterhouses and Small Scale Slaughterhouses by its Bacteriological Results:

Table 5.19, show that poultry samples, which produced in Gaza and Israel automatic slaughterhouses have lower contamination with *Salmonella* (4.2%) than others produced in Gaza small scale slaughterhouses (20.7%), the difference between the two groups is statistically significant (P=0.008).. Samples which produced in Gaza and Israel automatic slaughterhouses have higher contamination with total plate count more than 500000/g (8.3%), than others sample produced in Gaza small scale slaughterhouses (4.4%), the difference between the two groups is not statistical significant difference (P= 0.31).

Samples, which produced in Gaza and Israel automatic slaughterhouses, have lower contamination with *Staphylococcus aureus* > 50/g (4.2%), than others produced in Gaza small scale slaughterhouses (14.8%), the variations between the two groups is statistically significant (P= 0.05). In addition, samples which produced in Gaza and Israel automatic slaughterhouses have lower contamination with *E. coli* >100 /g (8.3%), than others produced in Gaza small scale slaughterhouses (31.1%), the difference between the two groups, is statistical significant difference (P=0.002).

Table 5.19: Distribution sample produced by automatic slaughterhouse address by its bacteriological results

Slaughterhouse Sample Address	Bacteriological Results				O.R.		P. Value
	Accept		Reject				
	No.	%	No.	%	Value	CI	
Salmonella							
Small Slaughterhouse	107	79.3	28	20.7	6.02	1.38-26.32	0.008*****
Automatic Slaughterhouse	46	95.8	2	4.2			
Total	153	83.6	30	16.4			
TPC							
Small Slaughterhouse	129	95.6	6	4.4	1.96	0.53-7.25	0.31
Automatic Slaughterhouse	44	91.7	4	8.3			
Total	173	94.5	4	5.5			
Staphylococcus aureus							
Small Slaughterhouse	115	85.2	20	14.8	0.25	0.06 -1.11	0.05*****
Automatic Slaughterhouse	46	95.8	2	4.2			
Total	161	88	22	12			
E. coli							
Small Slaughterhouse	93	68.9	42	31.1	0.2	0.07 – 0.6	0.002*****
Automatic Slaughterhouse	44	91.7	4	8.3			
Total	137	74.9	46	25.1			

*** Statistically significant**

Small-scale slaughterhouses had more poultry contaminated with *Salmonella, Staphylococcus aureus,* and *E. coli* than automatic slaughterhouses. That may be attributed to small slaughterhouses had less facilities, no or little application of GMP, workers were less experienced, had bad outdoor environment, practiced slaughtering of other poultry rather than chicken and turkey and their poultry not inspected by official organization, that lead to maximizing contamination.

Paradoxically, small scale slaughterhouses had lower (4.4%) contamination with TPC than others (8.3%). It may be because their samples were taken hot fresh, but the large scale slaughterhouses its samples were taken chilled or frozen, which were exposed to more handling, had more time between production and sampling and inadequate storage

temperature that aid in growth of Psychrophilic and Psychrotrophic bacteria (bacteria favour growth at temperatures less than 20 °C).

5.3.7 Relationships Between Bacteriological Analysis in Poultry and Presence of *Salmonella*:

Table 5.20, show that poultry samples, which have TPC equal, or less than $5x10^5$/g have less contaminated samples with *Salmonella* (16.2%) than others which have (>$5X10^5$/g) TPC (20%), the variations between the two groups are not statistically significant (P=0.75). Samples, which have higher contamination with *Staphylococcus aureus* (> 50/g) contaminated with *Salmonella* (31.8%), more than others which are with acceptable level (≤ 50/g) of *Staphylococcus aureus* (14.3%), the difference between the two groups are statistical significant (P= 0.037). In addition, samples which have higher contamination with *E. coli* (>100 /g) have contaminated with *Salmonella* (26.1%), more than others with acceptable contamination level (≤100/g) with *E. coli* (13.1%), the variations between the two groups is statistically significant (P=0.04).

Table 5.20: Relationships between bacteriological analysis in poultry and presence of *Salmonella*

Variable	*Salmonella* Results				O.R.		P. Value
	Positive		Negative				
	No.	%	No.	%	Value	CI	
TPC							
≤ 500000	28	16.2	145	83.8	0.77	0.16 – 3.83	0.75
>500000	2	20	8	80			
Total	30	16.4	153	83.6			
Staphylococcus aureus							
≤ 50	23	14.3	138	85.7	0.36	0.13 – 0.97	0.037*****
>50	7	31.8	15	68.2			
Total	30	16.4	153	83.6			
E. coli							
≤ 100	18	13.1	119	86.9	0.43	0.19 –0.98	0.04*****
>100	12	26.1	34	73.9			
Total	30	16.4	153	83.6			

*** Statistically significant**

The present study found positive relationship between TPC (above acceptable level postulated in PS) and *Salmonella* presence but the relationship was not statistically significant. That can be due to TPC is considered as quantitative test (increase during inadequate handling) while *Salmonella* test is qualitative (present or not) and there are differences between reservoirs of each of them. This finding is in agreement with Castillo-Ayala, et al., 1993 where there was no relationship between TPC and *Salmonella* isolated from fresh chicken.

The study showed there was a statistical relation ship between the presence of *Salmonella* in poultry meat and contamination with *Staphylococcus areuse*, may be related to miss of workers handling as Michigan Department of Agriculture, 2005 mentioned that *Staphylococcus areuse* considers as an indicator of handling abuse.

The study showed there was a statistically significant positive relationship between *Salmonella* and *E. coli*, which may be due to both of *Salmonella* and *E. coli* are two

genuses related to Enterobacteriaceae family that have the same reservoir of intestinal tract of animals and humans.

In general, presence of *Salmonella* in poultry meat had been reported all over the world. In 1983 Kampelmacher found *Salmonella* in raw chickens in different countries was 13% in West Germany, 45% in USA, 35% in England and 73% in Netherlands. In 1996, Wilson, et al., found *Salmonella* in 7% of retail chickens in Northern Ireland, while Geilhausen, et al., found 20% of fresh chicken breast of German, Dutch and French origin had *Salmonella*. In 1999, *Salmonella* was found by Uyttendaele, et al., in 36.5% of poultry carcasses and poultry products of retail market in Belgium. In 2001, *Salmonella* was found by Murakami, et al., in 37.8% of samples from raw chicken parts in Western Japan and was found by Beli, et al., in 6.5% of chicken meat samples in Albania.

In 2002, Dominguez, et al., showed that 35.83% of samples from chicken meat for sale had *Salmonella* in Spain. In 2003, Tibaijuka, et al., detected *Salmonellae* in 12.3%, 53.1% and 28%, with mean rate of about 18%, of chicken meat, gizzard and liver samples respectively in Addis Ababa.

5.4 Workers Attitude to Improve Poultry Production

During qualitative data collection, the study population were asked, on site, about their suggestions for improving branch of poultry processing and marketing. Their answers can be summarised in the following points:

1. Organizing local market, regarding selling of slaughtered poultry through changing the way of production and marketing to be depend on automatic poultry slaughterhouses to supply chilled or frozen poultry to shops which sale that products, and stopping slaughtering poultry in small places which sale fresh un-chilled poultry.

2. Keeping temperature stable during production, transporting, storing, and selling poultry meat.

3. Improving the quality of the products and packaging.

4. They need credit supplier as approval sources of live poultry to fix the price and profits.

5. They need of veterinary care in the farm for live poultry to assuring the safety and inspection before selling.

6. Raising awareness of farmers regarding proper breeding methods, specially delaying marketing live poultry after medications to safe of drug residue.

7. Poultry meat processing and selling shops should be licensed and under supervision.

8. Supervising eggs intended for hatching to be sure that are safe for producing chicks free of *Salmonella*.

Chapter 6

Conclusion and Recommendations

CHAPTER 6

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

Food-borne diseases take a heavy toll of human life and cause a great deal of suffering. *Salmonella* is considered one food-born disease, and poultry is one of the main *Salmonella* sources. This cross sectional study design conducted to identify *Salmonella* prevalence in fresh, chilled, and frozen poultry (chicken and turkey). Poultry produced by licensed sites by Gaza Municipality, 32 small scale slaughterhouses; only large slaughterhouse in Gaza and two companies importing poultry from Israel. Data was collected through direct interview through structured questionnaire; samples of poultry (183) were collected and examined in the MOH Public Health Laboratory. The study demonstrated the following results:

6.1.1. Workers Characteristics:

All workers were males older than 17 years old and 77% of them had more than 6 years of education. Only 17% of them did not pass medical examination. Workers wearing clothes special for work were 31% and about 6% of all workers complied with washing hand regulations. Thirty one percent of workers heard about *Salmonella* while 23% of them knew poultry as a source of *Salmonella* that they recognized as causing of illness.

6.1.2. Workplaces:

All workplaces were connected to municipal water network and to sewage net and only 11% did not have adequate water supply. Only 11% of workplaces had acceptable outdoor environment conditions, while 43% of places had valid municipal license. Solid wastes generated by workplaces was removed by municipal workers for 74% of places and the rest removed by workers of workplaces, while 46% of places let blood to go to sewage net.

Work places having wire mesh network for preventing pests were 17% of places, and those employing chemical and mechanical means for controlling pests were 66% of whom 83% were employing chemicals and the rest using mechanical and UV light. Workplaces employing paste detergents for cleansing equipment were 57% of places, and 54% of them employing liquid soap. Work places using hot water for cleansing were 79% of places, but only 3% were provided with continuous source of hot water. Non of the places employed chlorine or any other chemical for the carcasses. Only 6% of places were selling poultry raised in Israel, while the remaining were selling poultry raised in Gaza Strip governorates.

6.1.3. Samples Results:

Samples positive for *Salmonella* were 3% for turkeys, 19% for chicken regardless of temperature treatment. Places that separate processing of frozen, chilled and fresh chicken and turkeys from slaughtering, defeathering and evisceration of live birds had less contamination than places without separation that made (31/33) 93.9% of places selling frozen, chilled and fresh. There was no great difference between fresh and chilled samples regarding *Salmonella* contamination as both had about 1/5 positives, but frozen samples were free. There was no relationship between *Salmonella* contamination and TPC number, but there was a relationship between *Salmonella* and *Staphylococcus aureus* and

Escherichia coli. Geographically, Shati Camp and Sheikh Redhwan had the highest contamination with *Salmonella*. There was a link between bad outdoor environment and contamination. Liquid detergents were more effective than paste detergents that led to higher contamination. Large scale slaughterhouses had lower contamination with *Salmonella*, *Staphylococcus aureus* and *E. coli* than small-scale places and the difference was statistically significant.

6.2 Recommendations

The findings of this study enabled the researcher to set the following helpful recommendations, that contributing to reduce *Salmonella* and others biological hazards in the poultry which considered economic health problem.

6.2.1 General Recommendations:

1. Imposing and enacting official regulations regarding poultry processing and selling places, which include:
    a) Improving indoor and outdoor environment conditions.
    b) Places should be provided with liquid detergents.
    c) Separating processing stages from live bards.
    d) Separating final products from processing stages.
    e) All poultry processing and selling places should have official licenses and are under periodic supervision by authorities.
    f) Obliging persons dealing with poultry selling to pass routine medical examinations and to increase their hygienic practices.

2. Launching health education campaign for food handlers and consumers about proper handling, preparing and storing of food, especially poultry.
3. Raising awareness of farmers regarding proper breeding methods so as to prevent Salmonellosis in poultry.

6.2.2 Recommendation for Decision Makers:

It is recommended that Decision makers informed to:

1. Establishing a central or large semi automated poultry slaughterhouses that comply with relevant modern techniques and standards including the HACCP, see flowchart of poultry processing in Gaza City Slaughterhouses (annex 13) which reveal the variation between two processing methods.
2. Change the small-scale shops of slaughtering and processing poultry into just selling chilled or/and frozen poultry only.
3. Establishing a programme for continuous surveillance of *Salmonella* and all other food-born pathogens within food items.

6.2.3 Research recommendations:

1. Other similar studies including West Bank and Gaza Strip to provide national and seasonal data regarding *Salmonella* in poultry meat.
2. Further studies on national level to identify *Salmonella* serotypes prevalent in Palestine.

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Personal Communication

Saed Seyam Director of Veterinary Services of MOA at Crossing Points (July2005): Breeding Poultry in Gaza Strip. Personal Communication.

Appendices

Annex 1

Approval of General Director of Primary Health Care in Gaza Strip to Analyse the Samples of the Study in the Public Health Laboratory

Palestinian National Authority
Ministry Of Health

السلطة الوطنية الفلسطينية
وزارة الصحة

الرقم : ٩٠ ٨٧
التاريخ: 30/ 4/ 2005

السيد / مدير عـــــام الرعاية الأولية المحترم

تحيه طيبة وبعد ,,,,,

الموضوع : دراسة حول مدى انتشار السالمونيلا فى الدواجن بمدينة غزة

برجاء التكرم بالعلم أنني سأقوم بعمل دراسة حول مدى انتشار الســـالمونيلا فـــى لحوم الدواجن فى مدينة غزة وهذه الدراسة ضمن متطلبات دراسة الماجستير فى الصحـــة العامة فى جامعة القدس .

برجاء إجراءات سيادتكم بالموافقة لى بإجراء الفحص المخبرى الميكروبـــى فـــى مختبر الصحة العامة .

وتفضلوا بقبول فائق الاحترام .....

رئيس قسم مراقبة الأغذية

محمود حميد

Annex 2

Helsinki Committee Approval

بسم الله الرحمن الرحيم

Palestinian National Authority
Ministry of Health
Helsinki Committee

السلطة الوطنية الفلسطينية
وزارة الصحة
لجنة هلسنكي

Date: 3/5/2005

التاريخ: 2005/5/3

Mr./ Mahmoud Humaid

السيد: محمود حميد

I would like to inform you that the committee has discussed your application about:

نفيدكم علماً بأن اللجنة قد ناقشت مقترح دراستكم حول:-

Prevalence of Salmonella in Poultry Meat in Gaza City, 2005.

دراسة انتشار ميكروب السالمونيلا في الدواجن في مدينة غزة.

In its meeting on May 2005
and decided the Following:-
To approve the above mention research study.

و ذلك في جلستها المنعقدة لشهر مايو 2005
و قد قررت ما يلي:-
الموافقة على البحث المذكور عاليه.

Ministry of Health
Helsinki Committee

Signature
توقيع

Member
عضو

Member
عضو

Chairperson

Conditions:-

- Valid for 2 years from the date of approval to start.
- It is necessary to notify the committee in any change in the admitted study protocol.
- The committee appreciate receiving one copy of your final research when it is completed.

Gaza Etwam – Telefax 972-7-2878166

Annex 3

Formed Consent (Arabic Version)

الرقم: --------

التاريخ: / /2005

عنوان البحث

مدى انتشار ميكروب السالمونيلا في لحوم الدواجن

في مدينة غزة 2005

الأخ الكريم:

صاحب/مسئول مذبح أو محل ذبح وتنظيف الدواجن نود أن نخبركم أننا سوف نقوم بعمل دراسة بحثية خاصة لبرنامج ماجستير الصحة العامة في جامعة القدس حول مدى انتشار ميكروب السالمونيلا في الدواجن.المجهزة بعد ذبحها " المبردة / المجمدة " في منطقة مدينة غزة.

وقد وقع الاختيار على محلكم/مذبحكم للمشاركة في تزويدنا بالمعلومات اللازمة لتعبئة الاستبانة ومصدرا لعينات لحم الدواجن التي سيتم فحصها مخبريا, ونعلمك أن المعلومات التي سيتم الحصول عليها هي خاصة بالبحث فقط وستكون في سرية كاملة وستستعمل لتقييم انتشار ميكروب السالمونيلا في الدواجن بعد ذبحه وتجهيزه للبيع.

إن مشاركتكم في هذا البحث لن تلحق بكم أي ضرر وسيقدم أفضلية نحو المعرفة وتقييم الوضع الأمر الذي سيقدم النفع للصالح العام في المستقبل من الناحية الصحية والاقتصادية.

ولكم جزيل الشكر والتقدير.

الباحث /محمود حميد

غزة – الدرج عيادة الصوراني ت /2801323

إقرار بالموافقة

التاريخ: / /2005

أنا الموقع أدناه/----------.

وصاحب/ مسئول مذبح الدواجن في مدينة غزة/ ---------- لقد تفهمت البحث ولا مانع لدي من الإجابة علي كافة أسئلة الاستبيان.

كذلك أعلم وأفهم أن معلومات البحث ستكون محل السرية التامة ولن يتم استعمالها إلا بهدف المعرفة العلمية البحثية لغرض التخطيط الصحي والاقتصادي.

Annex 4

Formed Consent

Number: ----------
Date: / / 2005

Research Title: Prevalence of *Salmonella* in Poultry meat in Gaza city 2005

Dear Owner/Director,

Kindly I would like to inform you that you have been selected to be part of my research study "Prevalence of Salmonella in Poultry meat in Gaza city 2005' as part of the requirement for Master degree Program organized by Al-Quds University- Public health Program. Your facility has been thoroughly selected as a source of data by filling a well and comprehensive a questionnaire for that purpose.

All the information given from your side is top confidential and will be used to evaluate the prevalence of *Salmonella* in Poultry meat handling and marketing. Your participation is greatly appreciated and no information given would be used against you whatsoever.

Thanking you in advance for your cooperation.

Best Regards.

The researcher

Mahmoud A. Humaid

Date: / / 2005

I, the undersigned,…….. in my capacity as owner/Director for poultry slaughtering in Gaza City, completely understands the objectives of this research and has the full desire to fill in the following questionnaire.

As well as, I do realize that all information given will be top confidential and will be used for research purposes and health and economic planning.

Annex 5

الرقم: ----------

التاريخ: ----------

أولا: بيانات شخصية عن صاحب المحل أو المسئول فيه:

1) العمر: **----------**

2) الجنس: ☐ **ذكر,** ☐ **أنثى**

3) عنوان السكن: ☐ **(مدينة غزة),** ☐ **(شمال غزة),** ☐ **(الوسطى),** ☐ **(خانيونس),** ☐ **(رفح)**

4) عدد سنوات التعليم: **----------**

5) عدد سنوات العمل في مجال ذبح/تنظيف/الدواجن أو بيع لحومها: **----------**

ثانيا الحالة الصحية وممارسات العاملين

6) هل تجري فحص طبي دوري؟ 1أ **(نعم),** 2 **(لا)**

7) إذا كانت الإجابة نعم فهل طلب منك أخذ علاج نتيجة لما أظهرته النتيجة؟ 1 **(نعم),** 2 **(لا)**

8) في حالة إصابتك بمرض معدي كالانفلونزا, الرشح والحمى فهل؟

تبقى في بيتك حتى تشفى3 تغير مكان عملك, 2 تبقى في عملك, 1

9) إذا أصبت بجرح أو بدمل في جسمك فماذا تعمل؟

تبقى في البيت4 تضمد الجرح أو الإصابة بعصبة كاتمة, 3 تغير مكان عملك, 2 تستمر في عملك, 1

10) هل تقوم بتقليم أظافرك باستمرار؟ 1 **(نعم),** 2 **(لا),** 3 **(أحيانا)**

11) هل تغسل يديك بالماء والصابون خلال العمل؟ 1 **(نعم),** 2 **(لا)**

12) إذا كانت الإجابة نعم ففي أي من الحالات الآتية تغسل يديك بالماء والصابون؟

بعد الأكل2 بعد الخروج من المرحاض 1

بعد تفريغ أو ترحيل النفايات4 بعد التدخين 3

عند بدء العمل6 بعد ملامسة الملابس أو الجلد والشعر 5

في كل ما سبق8 بعد الانتهاء من العمل 7

13) هل تلبس زي خاص أثناء العمل؟ 1 **(نعم),** 2 **(لا)**

14) هل تعود إلى بيتك مرتديا نفس ملابس العمل؟ 1 **(نعم),** 2 **(لا)**

15) هل تلبس (قفازات) كفات الأيدي أثناء العمل؟ 1 **(نعم),** 2 **(لا)**

16) هل تلبس خواتم أو مجوهرات في يديك أثناء العمل؟ 1 **(نعم),** 2 **(لا)**

ثالثا: الثقافة والمعرفة بالسالمونيلا:

17) هل سمعت عن ميكروب السالمونيلا؟ ☐ 1 **(نعم),** ☐ 2 **(لا)**

18) في حالة الإجابة بنعم فهل سمعت ذلك خلال:

(أخرى) ☐ 4 (إرشاد), ☐ 3 (دورة تدريبية), ☐ 2 (التعليم/الدراسة), ☐ 1

19) ميكروب السالمونيلا هو:

(لا أعرف) ☐ 5 (فيروس), ☐ 4 (بكتيريا), ☐ 3 (خميرة), ☐ 2 (فطر), ☐ 1

20) هل تعلم ان ميكروب السالمونيلا يمكن ان يحدث مرض للإنسان؟ ☐ 1 **(نعم),** ☐ 2 **(لا),** ☐ 3 **(لا اعرف)**

21) هل تعلم ان الدواجن يمكن أن يكون مصدرا حاملاً لميكروب السالمونيلا؟

(لا اعرف) ☐ 3 (لا), ☐ 2 (نعم), ☐ 1

22) إذا كانت الدواجن حاملاً للسالمونيلا فما هو العضو الذي يعتبر مخزن للميكروب في الدواجن؟

(لا اعرف) ☐ 3 (الأمعاء), ☐ 2 (الأجزاء التي تؤكل), ☐ 1

23) هل سبق أن أصبت بمرض السالمونيلا؟ ☐ 1 **(نعم),** ☐ 2 **(لا),** ☐ 3 **(لا أعرف)**

24) هل سبق أن أصيب أحد الأقارب أو الأصدقاء أو العاملين معك أو الجيران بمرض السالمونيلا؟

(لا أعرف) ☐ 3 (لا), ☐ 2 (نعم), ☐ 1

25) إذا كانت الإجابة نعم فهل هو من:

(أخرى حدد) ☐ 7 (جار), ☐ 6 (صديق), ☐ 5 (العاملين معك), ☐ 4 (الإخوة), ☐ 3 (الأبناء), ☐ 2 (الوالدين), ☐ 1

رابعا: بيانات خاصة بالمذبح / المحل والخدمات فيه:

26) عنوان المحل:

الشيخ رضوان)[8] الشاطىء, [7] الرمال, [6] الصبرة, [5] الزيتون, [4] التفاح, [3] االدرج, [2] الشجاعية, [1]

27) هل المحل ترخيصه ساري المفعول؟ [1] **(نعم),** [2] **(لا)**

28) إذا كان الجواب لا فما هو تاريخ سريان آخر تجديد؟ **----------**

29) الجهات التي يتم أخذ موافقتها للترخيص هي:

(أخرى حدد)[6] (الدفاع المدني), [5] (الزراعة), [4] (الاقتصاد الوطني), [3] (الصحة), [2] (البلدية), [1]

30) الجهات الرسمية التي تفتش على محلكم؟ [1] **(البلدية),** [2] **(الصحة),** [3] **(الاقتصاد الوطني),** [5] **(الدفاع المدني)**

31) البيئة المحيطة بالمبنى: [1] **(جيدة),** [2] **(مقبولة),** [3] **(غير مقبولة)**

32) إذا كانت غير مقبولة فما هو السبب؟ **----------**

33) مساحة المحل: [1] **(كافية),** [2] **(غير كافية)**

34) نوع سقف المبنى: [1] **(باطون),** [2] **(أسبست),** [3] **(زينكو),** [4] **(أخرى)**

35) عدد العاملين: **----------**

36) أعمار العاملين بالسنة: **( ), ( ), ( ), ( ), ( ), ( )**

37) ما هي الدواجن التي يتم ذبحها/بيعها في محلكم ؟

بيع لحوم مبردة/مجمدة [4] **(أخرى),** [3] **(حبش),** [2] **(دجاج),** [1]

38) هل يتم الكشف على الدواجن المراد ذبحها؟ [1] **(نعم),** [2] **(لا)**

39) إذا كانت الإجابة نعم فأي الوزارات تقوم بالكشف عليها؟ [1] **(الزراعة),** [2] **(الصحة),** [3] **(الاقتصاد الوطني)**

40) هل يتم عزل الدواجن المريضة؟ [1] **(نعم),** [2] **(لا)**

41) كيف يتم التعامل الدواجن المريضة؟ [1] **(ارتجاعها لصاحبها),** [2] **(رميها)**

42) مصدر المياه: [1] **(بلدية),** [2] **(بئر خاص),** [3] **(أخرى حدد)**

43) إذا كان مصدر المياه ليس من البلدية فهل يضاف إلى الماء في المصدر مادة مطهرة كالكلور؟

(لا اعرف)[3] (لا), [2] (نعم), [1]

44) توفر المياه في المحل: [1] **(متوفرة يوميا ودائما),** [2] **(غير متوفرة بصورة دائمة)**

45) هل تقوم بتخزين احتياطي للمياه؟ [1] **(نعم),** [2] **(لا)**

46) إذا كان الجواب نعم فما هو نوع خزان المياه؟ [1] **(بلاستيك),** [2] **(حديد)**

47) وهل خزان المياه محكم الإغلاق؟ [1] **(نعم),** [2] **(لا)**

48) أرضية المبنى: 1 **(ناعمة وسهلة التنظيف),** 2 **(غير ذلك/غير ناعمة أوبها شقوق وكسور)**

49) الصرف الصحي في المحل: 1 **(متصل بشبكة عامة),** 2 **(متصل بحفرة امتصاصية)**

50) هل تلقي نفايات المحل السائلة خارج خط الصرف الصحي/ على الأرض في محيط محلكم؟ 1 **(نعم),** 2 **(لا)**

51) نوع التهوية في المحل: 1 **(طبيعية),** 2 **(صناعية),** 3 **(طبيعية وصناعية)**

52) التهوية: 1 **(جيدة),** 2 **(مقبولة),** 3 **(غير كافية)**

53) نوع الإضاءة في المحل: 1 **(طبيعية),** 2 **(صناعية),** 3 **(طبيعية وصناعية)**

54) الإضاءة: 1 **(جيدة),** 2 **(مقبولة),** 3 **(غير كافية)**

55) هل محلكم محصن ضد الحشرات والقوارض (الآفات)؟: 1 **(نعم),** 2 **(لا)**

56) هل تستخدم طرق لمكافحة الآفات؟: 1 (نعم), 2 (لا)

57) إذا كانت الإجابة نعم فهل طريقة المكافحة؟: 1 (ميكانيكية), 2 (كيماوية), 3 (كهربائيةٍ)

58) إذا كانت طريقة المكافحة كيماوية فهل تحتفظ بمواد المكافحة في مكان خاص بعيدا عن مكان العمل؟

(لا)2 (نعم), 1

59) هل تستخدم منظفات ومطهرات في عملية تنظيف الأدوات؟: 1 (نعم), 2 (لا)

60) إذا كانت الإجابة نعم فما هي نوعية مواد التنظيف والتطهير المستخدمة؟

(أخرى حدد)4 (صابون سائل), 3 (معجون صابون), 2 (كلور), 1

61) هل يتم تنظيف طاولات العمل والأدوات والمعدات؟: 1 (نعم), 2 (لا)

62) إذا كانت الإجابة نعم فهل تستخدم المياه الساخنة في التنظيف؟: 1 (نعم), 2 (لا)

63) كم مرة يتم تنظيف طاولات العمل والأدوات والمعدات يوميا؟: 1 (مرة), 2 (مرتين), 3 (ثلاثة مرات فأكثر)

64) عملية إزالة ريش الدواجن تتم: 1 (يدويا), 2 (آليا), 3 (آليا اوتوماتيكيا)

65) عملية إزالة ريش الدواجن تعتمد على المياه: 1 (المياه الساخنة), 2 (مياه حنفية عادية), 3 (أخرى)

66) في حالة استخدام المياه الساخنة في إزالة الريش هل تقوم بضبط درجة حرارتها؟ 1 (نعم), 2 (لا)

67) يتم تجديد المياه الساخنة المستخدمة لإزالة الريش يوميا: 1 (مرة), 2 (مرتين), 3 (ثلاثة مرات فأكثر)

68) بعد إزالة الريش وجميع الأحشاء يتم تنظيف الذبيحة بواسطة:

(يستخدم الماء لشطف الدواجن لأكثر من مرة)2 (يستخدم الماء لشطف الدواجن لمرة واحدة), 1

69) هل يضاف مواد مطهرة كالكلور في ماء تنظيف الدواجن بعد ذبحها؟ 1 (نعم), 2 (لا)

70) هل تستخدمون الاشعاع في التعقيم عندكم؟ 1 (نعم), 2 (لا)

71) هل يباع في المحل لحمة دواجن طازج مبرد أو مجمد؟ 1 (نعم), 2 (لا)

72) إذا كان الجواب نعم فهل مكان العرض منفصل عن مكان حجز الدواجن الحية وتنظيفها؟ 1 (نعم), 2 (لا)

73) وما هي سعة ثلاجات التبريد عندكم؟ ----------

النفايات وترحيلها/ التخلص منها:

74) أقرب مجمع نفايات للبلدية يبعد عن محلكم بمسافة------- متر

75) التخلص بنقل مخلفات محلكم لمجمع نفايات البلدية يتم: 1 (بواسطتكم), 2 (بواسطة عمال البلدية)

76) الدم الناتج عن عملية الذبح يتم التخلص منه بواسطة

(أخرى حدد)3 (تجميعه وترحيله إلى مجمع النفايات), 2 (تحويله إلى المجاري العامة), 1

77) ترحيل النفايات من المحل يتم: 1 (يوميا), 2 (كل يومين), 3 (أخرى حدد)

78) ترحيل النفايات من مجمع نفايات البلدية القريب منكم يتم: 1 (يوميا), 2 (كل يومين), 3 (أخرى)

79) ما هي نوعية مواد التغليف؟ 1 (بلاستيك), 2 (ورق أو كرتون), 3 (أخرى حدد)

80) مصدر الدواجن, بضاعتكم: 1 (مدينة غزة), 2 (شمال غزة), 3 (الوسطى), 4 (الجنوب), 5 (أخرى)

الأسئلة ثلاث التاليات 81, 82, 83 خاصة بالمذابح الآلية الميكانيكية

81) هل مكان استقبال وحجز الدواجن الحية منفصل ومستقل عن المنتج المجهز بعد إزالة الريش: 1 (نعم), 2 (لا)

82) أقصى مدة لحجز الدواجن الحية في المذبح هو: 1 (يوم), 2 (أكثر من يوم)

83) هل يوجود مصدر دائم للماء الساخن؟ 1 (نعم), 2 (لا)

الأسئلة 84, 85, 86, 87, 88, 89, 90, 91, 92, 93 خاصة لمنتجي ومتداولي لحوم الدواجن المجمدة

84) هل يوجد لديكم ثلاجة لإجراء عملية تجميد لحوم الدواجن الطازجة؟ 1 (نعم), 2 (لا)

85) إذا كانت الإجابة نعم فما هي سعة الثلاجة (بالمتر المكعب)؟ ----------

86) وما هي درجة الحرارة المستخدمة للتجميد؟ ------ م°

87) هل يوجد لديكم ثلاجة لحفظ لحوم الدواجن المجمدة؟ 1 (نعم), 2 (لا)

88) إذا كانت الإجابة نعم فما هي سعة الثلاجة (بالمتر المكعب)؟ ----------

89) وما هي درجة الحرارة المستخدمة لحفظ لحوم الدواجن المجمدة؟ ------ م°

90) هل ثلاجات التبريد والحفظ مزودة بساعة لبيان درجة الحرارة؟ 1 (نعم), 2 (لا)

91) هل يتم تسجيل الحرارة لثلاجات التبريد والحفظ؟ 1 (نعم), 2 (لا)

92) إذا كانت الإجابة نعم فكيف يتم التسجيل؟ 1 يدويا, 2 أوتوماتيكي

93) هل تقومون بنقل اللحوم المبردة والمجمدة في سيارة مبردة ومخصصة للغرض؟ 1 (نعم), 2 (لا)

خامسا: بيانات خاصة بالعينات المأخوذة

94) نوع العينة 1 (لحم دجاج), 2 (لحم حبش), 3 (أحشاء دجاج), 4 (أحشاء حبش)

95) مصدرها 1 (مدينة غزة), 2 (شمال غزة), 3 (الوسطى), 4 (جنوب غزة), 5 (أخرى)

96) مكان إنتاجها 1 (نفس مكان أخذ العينة), 2 (إسرائيل), 3 (أخرى)

97) حالتها 1 طازجة, 2 مبردة مسبقا, 3 مجمدة

98) ما هو اقتراحاتكم لتحسين الوضع في فرع إنتاج وتسويق الدواجن؟

1-

2-

3-

- **تم إجراء المقابلة بمعرفة/**----------

مرفق نتيجة الفحص ألمخبري للعينات التي تم تحليلها: 1 (ايجابي), 2 (سلبي)

Annex 6

Questionnaire Number:----------

Date:----------

1st Personal details for the owner

1) Age: **--------**

2) Gender 1 **Male** 2 **Female**

3) Address :1 **Gaza** 2 **Northern Gaza** 3 **Middle zone** 4 **Khan Younis** 5 **Rafah**

4) Years of Education:**-------**

5) Years of occupation in slaughtering Poultry: **-------**

2nd.Health Status and practices of workers

6) Do you make a periodic routine medical examination: 1 **Yes** 2 **No**

7) If yes, has any medicine been prescribed? 1 **Yes** 2 **No**

8) In a case you have fever or flue, Do you

1 Stay at work 2 Change tasks 3 Stay at home till curing

9) In case of injuries or abscises:

1 **Stay at work** 2 **Change tasks** 3 **Close the wounds** 4 **Stay at home**

10) Do you always cut your nails? 1 **Yes** 2 **No** 3 **Sometimes**

11) Do you wash your hands with soap while working? 1 **Yes** 2 **No**

12) If yes, in which cases you wash your hands?

1 After toilette

2 After eating

3 After smoking

4 After discharging wastes

5 After touching clothe, skin and hair

6 When staring work

7 After work

8 All above cases

13) Do you wear special uniform during work? 1 **Yes** 2 **No**

14) Do you go back home wearing the working clothes? 1 **Yes** 2 **No**

15) Do you wear special gloves during work? 1 **Yes** 2 **No**

16) Do you wear any rings while working? 1 **Yes** 2 **No**

3rd background and knowledge about salmonella

17) Have you heard about salmonella Microbe? 1 **Yes** 2 **No**

18) If yes, does it through:

1 Education 2 Training 3 Workshops 4 Others

19) Is the salmonella;

1 A Mould 2 A Yeast 3 A Bacteria 4 A Virus 5 Do not know

20) Do you realize that salmonella would cause a disease to human?

1 **Yes** 2 **No** 3 **Do not know**

21) Do you know that poultry is a reservoir for salmonella?

1 Yes 2 No 3 Do not know

22) If yes, which is the origin in which salmonella has been concentrated?

1 Eatable parts 2 Intestine 3 Do not know

23) Have you got salmonella? 1 **Yes** 2 **No** 3 **Do not know**

24) Have any of your family, friends, neighbors and work mates got salmonella?

1 Yes 2 No 3 Do not know

25) If yes, please specify:

1 Parents 2 Sons 3 Brothers 4 Work mates 5 Friend 6 Neighbor 7 Others

4th. Slaughter Details

26) Address of slaughtering place:

1 **SHiggaea** 2 **Darag** 3 **Tofah** 4 **Ziton** 5 **Sabra** 6 **Remal** 7 **S. Camp** 8 **S. Rdwan**

27) Do you have a valid license 1 **Yes** 2 **No**

28) If no, what is the last license you had? ----------

29) What is the official organization you have to get a license from?

1 Municipality 2 MOH 3 MONE 4 MOA 5 Civil Defense 6 Others

30) Please specify the official organizations that run inspection at your facility? **----------**

31) Surrounding environment 1 **Good** 2 **Accepted** 3 **Not Accepted**

32) If not acceptable, what is the reason stand behind? **----------**

33) Area 1 **Sufficient** 2 **Insufficient**

34) Kind of the ceiling 1 **Concrete** 2 **Asbestos** 3 **Metal roof** 4 **Others**

35) Number of workers **----------**

36) Their ages: **( ) , ( ) , ( ) , ( ) , ( ) , ( )**

37) What kinds of Poultry slaughtered? 1 **Chicken** 2 **Turkey** 3 **Others**

38) Is there any inspection on the Poultry slaughtered? 1 **Yes** 2 **No**

39) If yes, please specify who make it?

40) Is there any isolation for infected poultry? 1 **Yes** 2 **No**

41) How do you handle with the infected poultry?

42) Source of water used? 1 **Municipality** 2 **Special well** 3 **Others**

43) If the water used is not from the municipality, Is there any chlorination for it?

1 Yes 2 no 3 don't know

44) The availability of water 1 **Always available** 2 **Not available all**

45) Do you have additional water reservoir? 1 **Yes** 2 **No**

46) If yes, what is the type of the water tank?**---------**

47) Is the additional water tank well closed 1 **Yes** 2 **No**

48) Floor 1 **Smooth and easily cleaned** 2 **Not smooth, cleavages, others**

49) Sewage 1 **Connected with the sewage net** 2 **Connected with absorb well**

50) Do you throw liquid wastes in the vicinity 1 **Yes** 2 **No**

51) Types of ventilation: 1 **natural ventilated** 2 **artificial** 3 **both**

52) Ventilation Condition: 1 **Good** 2 **Accepted** 3 **Not enough**

53) Lightening: 1 **Natural** 2 **Artificial** 3 **Both**

54) Lightening Condition: 1 **Good** 2 **Accepted** 3 **Not enough**

55) Is your place protected against pests? 1 **Yes** 2 **No**

56) Do you use any of known methods to prevent pests? 1 **Yes** 2 **No**

57) If yes, please specify whether it is: 1 **Mechanical** 2 **Chemical** 3 **Electricity.**

58) If you use chemical pesticides, do you keep it in a far safe place? 1 **Yes** 2 **No**

59) Do you use cleaning agents and disinfectants? 1 **Yes** 2 **No**

60)If yes, Please specify their kind? 1 Chlorine 2 Paste soap 3 Liquid soap 4 Others

61)Do you always clean equipments of all kinds you use? 1 yes 2 no

62)If yes, do you use hot water? 1 Yes 2 No

63) How many times do you clean equipment daily? 1 Once 2 Twice 3 Others

64)How do you make feathering? 1 Manually 2 Mechanical 3 Automatically

65)Is the feathering done by using 1 Hot water 2 Tap water 3 Others

66) In case of hot water, do you control its temperature? 1 Yes 2 No

67)Renewing hot water used in feathering daily 1 Once 2 Twice 3 Three or more

68)After feathering and intestine removal, how do you clean the carcass?

1 By running water 2 Using the same water for several carcasses.

69)Is there any additional disinfectants, as chlorine, to cleaning water 1 yes 2 No

70)Do you use irradiation as sterilizing agent? 1 Yes 2 No

71)Do you sell chilled or frozen poultry? 1 Yes 2 No

72)If yes, do you sell them in a separate area? 1 Yes 2 No

73)What is the capacity of the refrigerators? ----------

Wastes and the means of discharge

74) What is the nearest municipality garbage collection? ----------

75)Is waste discharge from your facility done by 1 You 2 Municipality workers

76)How do get rid of blood

1 To the sewage net 2 Collected and discharged to the nearest garbage 3 Others

77)waste discharge is done 1 Daily 2 Every two days 3 Others

78) discharging of the wastes from the nearest municipality garbage collection is done

1 Daily 2 Every two days 3 Others

79)Types of packaging material 1 Plastics 2 Paper sheets 3 Others

80)Source of poultry 1 Gaza 2 Northern Gaza 3 Middle zone 4 South Gaza 5 Others

Questions No. 81,82 and 83 is related to automatic slaughtering

81)Is the reception and holding of a live poultry segregate from that for those cleaned and

ready to deliver? 1 Yes 2 No

82)What is the maximum period for holding alive poultry? 1 A day 2 More than day

83)Do you have permanent source for hot water 1 Yes 2 No

Questions from No.84 to 93 are related to the handlers of frozen poultry.

84)Do you have a freezer for processing 1 Yes 2 No

85)If yes, what is the capacity? ----------m^3

86)Temperature used for freezing. ----------m^3

87) Do you have freezer for storage 1 Yes 2 No

88)If yes, what's its capacity? ----------m^3

89)Temperature used in storage? ----------°C

90)Are the freezers and refrigerators having temperature indicators? 1 Yes 2 No

91)Do you make any records for temperatures? 1 Yes 2 No

92)If yes, how do you record them? 1 Manual 2 Automatic

93)Do you transport chilled and frozen meat in a special vehicle? 1 Yes 2 No

5th.Samples details:

94)Sample types 1 Chicken 2 Turkey 3 Chicken offal 4 Turkey offal

95) Source of sample 1 Gaza 2 Northern Gaza 3 Middle zone 4 South Gaza 5 Others

96) Place of production 1 In site 2 Israel 3 Others

97)Status of sample 1 Fresh 2 Pre chilled 3 Frozen

98)What are your suggestions to improve production and marketing of poultry?

1-

2-

3-

The meting don by: ----------

Attach the lab analysis: ☐1 Positive ☐2 Negative.

Annex 7

Media

Nutrient Agar (Difco) for Heterotrophic Plate Count (H.P.C)

The typical formula (g/l):

Bacto beef extract, 3.0; Bacto peptone, 5.0; Sodium chloride, 8.0 and Bacto agar, 15.0.

31 grams of the powder were suspended in one liter of distilled water, brought to boil to dissolve completely, and sterilized by autoclaving at 121 °C for 15 minutes. The final pH was adjusted to 7.3 ± 0.1 at 25 °C.

Peptone Water (Oxoid) for Dilution

The typical formula (g/l):
Peptone, 10.0 and Sodium chloride, 5.0.
15 grams of Peptone water were dissolved in one liter of distilled water, mixed well and distributed into final containers. The media was sterilized at 121 °C for 15 minutes. The final pH was adjusted to 7.2 ± 0.2.

Bacto Violet Red Bile Agar (Difco) *E. coli*

The typical formula (g/l):

Bacto Yeast extract, 3.0; Bacto Peptone, 7.0; Bacto No. 3, 1.5; Bacto Lactose, 10.0; Sodium chloride, 5.0; Bacto Agar, 15.0; Neutral red and 0.03; Bacto Crystal violet, 0.002.
41.5 grams of the powder were suspended in one liter distilled water or deionized water and heated to boiling to dissolve completely. Media was not autoclaved. The final pH was adjusted to 7.4 ± 0.2 at 25 °C.

Bacto Baird Parker Agar (Difco) for *Staphylococcus aureus*.
The typical formula (g/l):
Bacto Tryptone, 10.0; Bacto Beef extract, 5.0; Bacto Yeast extract, 1.0; Glycine, 12.0; Sodium pyruvate, 10.0; Lithium chloride, 5.0 and Bacto agar, 20.0.
63 grams of the powder were suspended in 950 ml distilled or deionized water, heated to boiling to dissolve completely, and sterilized by autoclaving at 121 °C for 15 minutes. Cooling to 45 – 50 °C, meanwhile, warm (45 - 50 °C) Bacto Egg Yolk Tellurite enrichment

supplement was added and mixed well with the prepared base. The final pH was adjusted to 7.0 ± 0.1 at 25 °C.

Lactose Broth (Difco) for *Salmonella* Pre Enrichment.

The typical formula (g/l):
Bacto beef extract, 3.0; Bacto peptone, 5.0 and Bacto lactose, 5.0.
13 grams of the powder were dissolved in one liter of distilled water. The media was sterilized at 121 °C for 15 minutes. The final pH was adjusted to 6.9 ± 0.2 at 25 °C.

Selenite F- Broth (Difco) for *Salmonella,* Enrichment.

The typical formula (g/l):
Bacto tryptone, 5; Bacto lactose, 4.0; Disodium phosphate, 10.0; Sodium acid selenite, 4.0 and L – cystine, 0.01.
23 grams of the powder were dissolved in one liter of distilled water and heated to boil. The media without sterilization, was distributed into sterile cups to a depth at least 5 cm. The final pH was adjusted to 7.0 ± 0.2 at 25 °C.

S.S Agar (Difco) for *Salmonella*.

The typical formula (g/l):
Bacto beef extract, 5.0; Proteose peptone, Difco, 5.0; Bacto lactose, 10.0; Bacto bile salts No. 3, 8.5; Sodium citrate, 8.5; Sodium thiosulfate, 8.5; Ferric citrate, 1.0; Bacto agar, 13.5; Bacto brilliant green, 0.00033; and Bacto neutral red, 0.025.
60 grams of the powder were suspended in one liter of distilled water and boiled for 2 – 3 minutes with frequent and careful swirling for complete dissolving. Media was not sterilized, after cooling to 55 – 60 °C; the media was distributed into sterile Petri dishes. The final pH was adjusted to 7.0°C ± 0.2 at 25 °C.

Bacto Bismuth Sulfite Agar (Difco) for *Salmonella*.

The typical formula (g/l):
Bacto beef extract, 5.0; Bacto peptone, 10.0 ; Bacto dextrose, 5.0; Disodium phosphate, 4.0; Ferrous sulfate, 0.3; Bismuth sulfite indicator, 8.0; Bacto agar, 20.0 and Bacto brilliant green, 0.025. 52 grams of the powder were dissolved in one liter of distilled water and boiled. The media is not autoclavaible. The final pH was adjusted to 7.7 ± 0.2 at 25 °C.

Bacto Xylose Lysine Desoxycholate (XLD) Agar (Difco) for *Salmonella*.

The typical formula (g/l):

Bacto yeast extract, 3.0; L-lysine, 5.0; Bacto xylose, 3.75; Bacto lactose, 7.5; Bacto saccharose, 7.5; Sodium desoxycholate, 2.5; Ferric ammonium citrate, 0.8; Sodium thiosulfate, 6.8; Sodium chloride, 5.0; Bacto agar, 15.0 and Bacto phenol red, 0.08.
57 grams of the powder were suspended in one liter of distilled water and the mixture was boiled to dissolve completely. Media was not autoclaved. The final pH was adjusted to 7.4 ± 0.2 at 25 °C.

Bacto Triple Sugar Iron (TSI) Agar (Difco) for Identification.

The typical formula (g/l):

Bacto beef extract, 3.0; Bacto yeast extract, 3.0; Bacto peptone, 15.0; Proteose peptone, Difco, 5.0; Bacto dextrose, 1.0; Bacto lactose, 10.0; Bacto sucrose, 10.0; Ferrous sulfate, 0.2; Sodium chloride, 5.0; Sodium thiosulfate, 0.3; Bacto agar 12.0 and Bacto phenol red, 0.024.
65 grams of the powder were suspended in one liter of distilled water, boiled to dissolve completely. Media was dispensed into tubes and sterilized in the autoclave for 15 minutes at 121° C. The final pH was adjusted to 7.4 ± 0.2 at 25 °C.

Lysine Iron Agar (Difco) for Identification.

The typical formula (g/l):
Bacto peptone, 5.0; Bacto yeast extract, 3.0; Bacto dextrose, 1.0; L. lysine hydrochloride, 10.0; Ferric ammonium citrate, 0.5; Sodium thiosulfate, 0.04; Bacto bromo cresol purple, 0.02 and Bacto agar, 15.0.
34.5 grams of the powder were suspended in one liter of distilled water, boiled to dissolve completely. Media was dispensed into tubes and sterilized in the autoclave for 15 minutes at 121 °C. The final pH was adjusted to 6.7 ± 0.2 at 25 °C.

Biochemical Tests
The Analytical Profile Index (API) 20 E and API Staph. strips (Bio Merieux) were used as the biochemical systems for identification of Gram-negative rod bacteria and Gram positive Cocci, respectively. The API 20 E and API Staph. strip consist of 20 micro-tubes containing dehydrated substrates. These tests were inoculated with bacterial suspensions, which reconstitute the media. The strips were incubated for 18 to 24 hours at 37 °C. During the incubation, metabolism produces changes that are either spontaneous or revealed by the addition of reagents. The standards were scored according to a reading table and the identification was obtained by referring to the API catalogue (BioMerieux).

Annex 8

Bacteriological Testes and Procedures

Bacteriological Procedures are Including the Following Testes:

Aerobic Plate Count (APC)

Decimal dilutions (10^{-2}, 10^{-3}, and 10^{-4}) of poultry meat samples (prepared as described in preparation of the sample) were used. One ml of each dilution was transferred aseptically into separate, Petri dishes in duplicate. 12-15 ml of melted (and cooled to 45 $^{\circ}$C) count agar was added to each plate. The plates were swirled gently and after the agar medium solidified, the plates were incubated for 48±2 h at 35 °C.

Isolation of Coliform Group Bacteria

1- Prepare Violet Red Bile Agar (VRBA) and pasteurize it by boiling for 2 min on day of use.

Homogenize 25 g sample at high speed for 1 min in 225 ml 0.1% peptone water. Prepare serial tenfold dilution in butter field's diluents or 0.1% peptone water in accordance with anticipated level of coliforms. Transfer two 1 ml aliquots of each dilution to Petri dishes.

2- Use either of two plating method for conventional method pour 10ml VRBA tempered to 48^{o}C into plates. Swirl plates to mix and let solidify.

Note: To prevent surface growth and spreading of colonies, overlay with 5 ml VRBA, and let solidify. If resuscitation is necessary, pour basal layer of 8-10 ml of tryptic soy agar tempered to 48^{o}C. Swirl plates to mix, and incubate at room temperature for 2 ± 0.5h. Then overlay with 8-10 ml of melted, cooled VRBA and let solidify.

Isolation of *E. coli*

To find *E. coli* among coliforms, use 100µg 4 methyl- umbelliferyl- beta-D- glucuronide (MUG) per 1 ml in the VRBA overlay and observe for fluorescent colonies under long wave UV light.

Isolation of *Staphylococcus aureus*

Decimal dilutions (10^{-2}, 10^{-3}, and 10^{-4}) of poultry meat samples (prepared as described in Preparation of the sample) were used. One ml of each dilution was transferred aseptically onto the surface of Baird Parker agar and was spread, using sterile bent glass-streaking rod. Plates were incubated for 45-48 h at 35 ºC. Typical *Staphylococcus aureus* colonies appeared as circular, smooth, convex, moist, 2-3 mm in diameter on un-crowded plates, gray to jet-black, frequently with light-coloured margin, surrounded by opaque zone and frequently with an outer clear zone. *S. aureus* is Coagulase and Catalase positive.
Suspected colonies were stained with Gram staining, *S.aureus* is Gram-positive cocci. Test

by Latex agglutination test, was also used for further confirmation **(FDA, 1995).**

Isolation of *Salmonella*

a- Pre-Enrichment

25 g sample was suspended in 225ml. Sterile lactose broth and blended for 2 min.

Homogenized mixture was aseptically transferred to sterile wide-mouth, screw-cap jar (500

ml), and let stand 60 min at room temperature. Sample mixtures were incubated for 24 ± 2

h at 35 °C.

b- Enrichment

One ml pre-enrichment mixture was transferred to 10 ml Selenite Cystine (SC) broth and another 1 ml to 10 ml Tetrathionate broth (TT). SC and TT broth were incubated for 24 ± 2 h at 35 °C.

Isolation and Identification

Enrichment broths were used to streak on Bismuth Sulfite (BS) agar, Xylose Lysine Desoxycholate (XLD) agar and *Salmonella-Shigella* (S.S) agar. Plates were incubated for 24 ± 2 h at 35 °C.

Plates were examined for presence of colonies that may be *Salmonella*.

Xylose Lysine Desoxycholate (XLD) agar: Pink colonies with or without black centres. Bismuth Sulfite (BS) agar: Brown, gray, or black colonies; sometimes they have a metallic sheen. *Salmonella-Shigella* (S.S) agar: Black colonies.

Confirmation Test

Two or more typical colonies were transferred to Triple Sugar Iron agar (TSI) and Lysine Iron agar (LIA). TSI and LIA were incubated for 24 ± 2 h at 35 °C.

Salmonella in culture typically produces alkaline (red) slant and acid (yellow) butt, with or without production of H_2S (blackening of agar) in TSI. In LIA, only tubes with distinct yellow butts [acidic (negative) reaction] were considered.

c- Serological Test

1- Serological Polyvalent Flagellar (H) Test

Growth from each urease-negative TSI agar slant was inoculated into Trypticase Soy Tryptose (TST) broth and incubated for 24 ± 2 h at 35 °C. 2.5 ml formalinized

physiological saline solution was added to 5 ml of TST broth culture and tested with polyvalent flagellar (H) antisera. 0.5 ml of polyvalent flagellar (H) antisera was added to the mixture of TST broth in tubes. Agglutination in the tubes considered positive.

2- Serological Polyvalent Somatic (O) Test

One drop of *Salmonella* polyvalent somatic (O) antiserum was added to one drop of TST broth in Petri dishes and mixed by a wooden applicator. Agglutination considered a positive reaction (FDA, 1995).

2005/ / التاريخ: **Annex 9**

نموذج طلب وتسجيل فحص لحوم دواجن ميكروبياً

Request and Registration of Microbiological Analyses for Poultry

Questioner No.الإستبانة…… …… …… الرقم

Sampling Dateتاريخ أخذ العينة .....

Substance Nameاسم المادة:……

Producer name & Addressاسم المنتج وعنوانه……

Name & Address of Shopper manاسم صاحب البضاعة وعنوانه……

Reception Dateتاريخ استلام العينة ……

Production Dateتاريخ الإنتاج ……

Expireتاريخ الانتهاء……

Temp. at. Labدرجة الحرارة عند وصول المختبر……

Sample Wightكمية العينة ……

Collected byاسم آخذ العينة وتوقيعه……

ملاحظات:……

……

نتائج الفحص:

No.	Sample	State	Result				
			TBC	Stap.	Coli.	E. coli	Salm.

2 ملاحظات:

.....

.....

Annex 10

Palestinian National Authority
Ministry of Health
General Administration of P.H.C
Public Health Laboratory

السلطة الوطنية الفلسطينية
وزارة الصحة
الإدارة العامة للرعاية الأولية
مختبـــر الصحة العامـــة

FOOD MICROBIOLOGY REPORT تقرير فحص ميكروبيولوجي أغذية

Code No..... الرقم السري Substance name ..... اسم المادة :

Trade mark..... علامة المنتج Producer name & addres ..... اسم المنتج وعنوانه

Name & address of shopper man ..... اسم صاحب البضاعة وعنوانه

Date..... تاريخ استلام العينة Expire..... تاريخ الانتهاء Product date..... تاريخ الإنتاج

Temp. at. Lab..... درجة الحرارة عند وصول المختبر Temp..... درجة الحرارة عند أخذ العينة:

From total..... من اصل Qua. of sample..... كمية العينة

Collect. By ..... اسم آخذ العينة وجهة العمل

ملاحظات:.....

.....

اسم الفاحص:.....

نتائج الفحص :

No	Sample	T.B.C	Staph. aureus	Coliform	E.coli	Sal.	Mold & Yeast				
							Mold	Yeast	O. Lacts		
1											
2											
3											
4											
5											
6											
7											
8											
9											
10											

التاريخ:..... توقيع رئيس القسم:.....

مدير المختبر:.....

PLAC

☐ المختبر مسئول عن العينات الواردة للمختبر.
☐ لا يجوز إعادة إصدار التقرير إلا بموافقة خطية من مدير المختبر.
* المختبر حاصل على شهادة اعتماد المختبرات (PSI 17)

Annex 11

Palestinian National Authority
Ministry Of Agriculture
General administration of Veterinary Services & Animal Health
Telfax : 08-2840017

السـلطة الوطنيـة الفلسـطينية
وزارة الـزراعـة
الإدارة العامة للخدمات البيطرية وصـحة الحيوان
تلفاكس: 08-2840017

Veterinary certificate to accompany day old chicks transported between Gaza & West bank and vice versa via Israel

1- Identification **No: (5)**

Type and Species	Breed	Total number of birds
One day old chicks	Ross	12000

2- Origin of the products: **Gaza**

Name and address of exporter.....ARABIA CO.

Name and address of the hatchery.....ARABIA HATCHERY

3- Destination of the products:.....HEBRON

Name and address of consignee:.....H. TAMEMY.

4- Declaration

I, the undersigned official veterinary officer hereby certify that the said birds:-

A- The country is free from Avian Influenza and Salmonella pullorum.

B- The hatchery and farm of origin of the said birds in a radius of 10 Km. has been free from Newcastle disease during the last 12 months.

C- The day old chicks described above originate from breeding farm ,that are known to be free from ,Mycoplasma spps,Salmonella typhimrium, Sal enteritidis

D- The breeding flocks/hatchery from which the said hatching eggs/birds originated were free from clinical evidence of infectious diseases.

E- The said birds are vaccinated against Newcastle disease.

Exit gate : Montar

Dr S .R . Seyam
Name & title of official vet officer

Date : 21 / 7 /2005

Resept No:

Total sum:

Sig.....

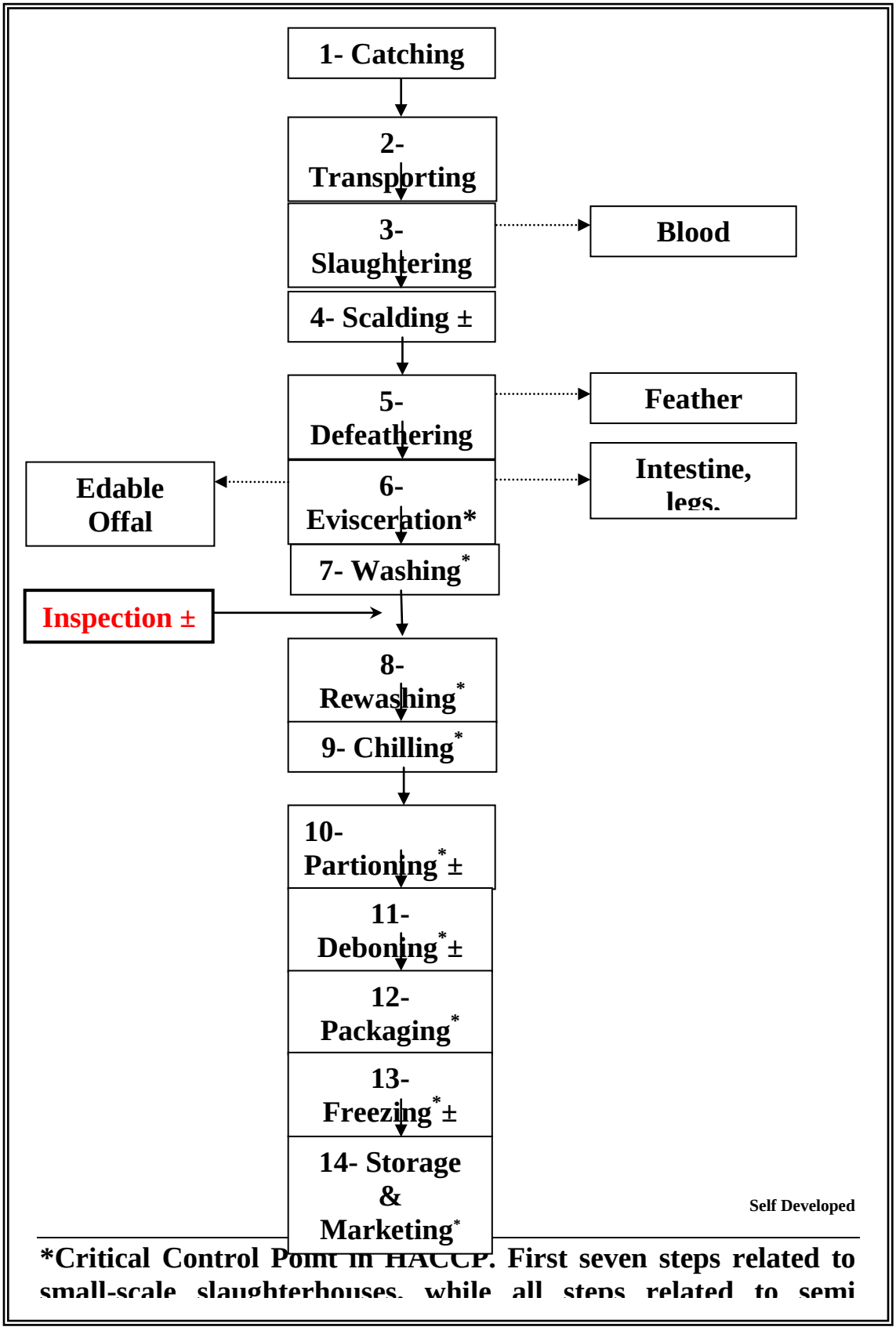
Official Stamp

Annex 12

Wash your Hands Frequently Throughout the Working Day

Annex 13

Flowchart of Poultry Processing in Gaza City Slaughterhouses



Self Developed

*Critical Control Point in HACCP. First seven steps related to small-scale slaughterhouses, while all steps related to semi

Annex 14

Distribution of Microbiological Result of the Study Samples

Sample No.	Microbiological Results			
	TPC	***Staph. aureus.***	***E .coli***	***Salmonella.***
1	20000	500	0	negative
2	2000	100	0	negative
3	1000	0	0	negative
4	200000	0	2000	negative
5	3000	0	0	negative
6	500	0	0	negative
7	3000	0	0	negative
8	0	0	0	negative
9	8000	0	0	negative
10	10000	0	0	negative
11	0	0	0	negative
12	0	0	0	negative
13	2500	0	0	negative
14	7000	0	0	negative
15	5000	0	0	negative
16	0	0	0	negative
17	200000	0	0	negative
18	3000	0	0	negative
19	40000	0	0	negative
20	10000	200	200	negative
21	30000	200	0	negative
22	700000	300	0	negative
23	8000	1500	0	negative
24	15000	0	0	negative
25	300000	0	0	positive
26	600000	1500	0	positive
27	400000	0	0	negative
28	600000	200	0	negative
29	200000	400	0	negative
30	600000	0	10000	negative
31	200000	200	40000	negative
32	10000	200	0	negative
33	30000	0	0	negative
34	10000	100	0	negative

35	10000	0	0	negative
36	40000	0	0	negative
37	60000	0	0	negative
38	100000	0	0	negative
39	300000	0	0	negative
40	60000	0	0	negative
41	20000	400	0	negative
42	30000	0	0	negative
43	80000	0	0	negative
44	70000	200	0	negative
45	3000	0	0	negative
46	20000	0	0	negative
47	300000	0	0	negative
48	50000	200	0	negative
49	10000	0	500	negative
50	6000	600	0	negative
51	40000	1000	3000	negative
52	60000	500	0	negative
53	100000	400	0	negative
54	20000	0	0	negative
55	30000	300	2000	negative
56	100000	0	200	positive
57	40000	0	0	negative
58	20000	0	0	negative
59	100000	500	500	negative
60	200000	2000	4000	negative
61	300000	2000	0	negative
62	150000	1000	0	negative
63	200000	2000	0	positive
64	400000	0	0	positive
65	60000	3000	0	negative
66	100000	500	0	negative
67	6000	0	0	negative
68	200000	0	0	negative
69	100000	0	0	positive
70	200000	1000	0	negative
71	30000	0	100	negative
72	60000	100	0	negative
73	10000	0	0	negative
74	10000	500	200	negative
75	20000	0	0	negative

76	150000	800	200	negative
77	10000	200	300	negative
78	10000	500	0	negative
79	20000	400	0	positive
80	30000	600	0	negative
81	20000	0	0	negative
82	150000	0	0	positive
83	30000	0	0	positive
84	1000	0	0	positive
85	8000	100	0	negative
86	5000	0	0	negative
87	20000	0	2500	negative
88	2000	500	0	negative
89	10000	0	0	negative
90	60000	1700	6000	positive
91	100000	300	0	positive
92	20000	0	5000	positive
93	20000	0	0	negative
94	3000	0	100	negative
95	6000	0	0	negative
96	9000	0	0	negative
97	300000	3000	10000	negative
98	500000	0	100000	negative
99	500000	800	100000	positive
100	800000	600	80000	negative
101	400000	300	6000	negative
102	300000	0	0	positive
103	800000	6000	200000	negative
104	1000000	5000	80000	negative
105	1000000	10000	100000	negative
106	300000	8000	10000	negative
107	40000	0	0	negative
108	10000	500	600	negative
109	3000	0	0	negative
110	10000	100	0	negative
111	40000	200	5000	negative
112	20000	0	0	negative
113	30000	200	0	negative
114	10000	0	0	negative
115	40000	0	2000	negative
116	8000	200	0	negative

117	500000	3000	0	negative
118	10000	2000	0	negative
119	100000	500	0	negative
120	300000	6000	40000	positive
121	100000	4000	0	negative
122	100000	8000	0	positive
123	60000	0	0	positive
124	100000	0	0	negative
125	200000	0	0	negative
126	200000	0	0	positive
127	300000	0	10000	positive
128	60000	0	5000	negative
129	100000	0	20000	negative
130	200000	0	0	negative
131	200000	400	0	negative
132	150000	200	0	negative
133	300000	300	2000	negative
134	10000	500	1000	negative
135	10000	600	0	positive
136	300000	0	0	positive
137	300000	0	0	positive
138	300000	0	0	negative
139	300000	1000	0	negative
140	200000	800	2000	positive
141	400000	5000	6000	positive
142	1000000	100000	20000	positive
143	10000	0	2000	positive
144	500000	200	5000	negative
145	30000	200	0	negative
146	100000	0	1000	negative
147	200000	1000	0	negative
148	120000	0	0	negative
149	100000	0	0	negative
150	200000	5000	0	negative
151	300000	0	0	positive
152	400000	0	0	negative
153	270000	0	500	positive
154	200000	0	0	negative
155	600000	0	0	negative
156	300000	0	6000	negative
157	270000	8000	0	negative

158	300000	10000	0	negative
159	3000	0	0	negative
160	400000	0	0	negative
161	200000	0	0	negative
162	100000	0	0	negative
163	200000	0	0	negative
164	40000	0	0	negative
165	20000	0	2000	negative
166	8000	0	0	negative
167	10000	0	0	negative
168	3000	0	0	negative
169	10000	0	0	negative
170	8000	0	0	negative
171	6000	0	0	negative
172	60000	800	300	negative
173	50000	0	200	positive
174	30000	0	0	negative
175	40000	100	300	negative
176	300	0	0	negative
177	500	0	0	negative
178	0	0	0	negative
179	0	0	0	negative
180	0	0	0	negative
181	0	0	0	negative
182	0	0	0	negative
183	1000	0	0	negative