## HYGIENE PRACTICES OF TABLE EGGS AT DIFFERENT WET MARKET IN SHER-E-BANGLA NAGAR AREA OF DHAKA NORTH CITY CORPORATION: A PUBLIC HEALTH CONCERN

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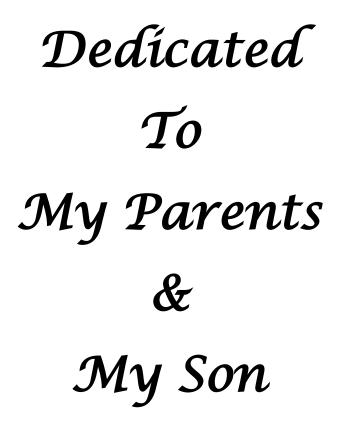
This is to certify that the thesis entitled, "HYGIENE PRACTICES OF TABLE EGGS AT DIFFERENT WET MARKET IN SHER-E-BANGLA NAGAR AREA OF DHAKA NORTH CITY CORPORATION: A PUBLIC HEALTH CONCERN" Submitted to the Department of Animal Nutrition, Genetics and Breeding, Faculty of Animal science and veterinary medicine, Sher-E-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of MASTER OF SCIENCE (MS) in Animal Nutrition embodies the result of a piece of bona fide research work carried out by JANNATUL MAWA, Registration No. 20-11091 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that such help or source of information, as has been availed of during the course of this investigation has been duly acknowledged by him.

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## LIST OF ACRONYMS AND ABBREVIATION

ABBREVIATION		FULL MEANING
ADB	=	Asian Development Bank
CDC	=	Center for Disease Control and Prevention
CFU	=	Colony Forming Unit
CONT'D	=	Continued
DGHS	=	Directorate General of Health
		Services
E. coli		Escherichia coli
e.g.	=	For example
et al.	=	And others/associates
FAO	=	Food and Agricultural
		Organization
FBD	=	Food Borne Disease
FDA	=	Food and Drug Administration
FSAI	=	The Food Safety Authority of Ireland
EU	=	European union
EMB	=	Eosin Methylene Blue Agar
ECDC		The European Centers for Disease Prevention
EFSA	=	The European Food Safety Authority
GoB	=	Government of Bangladesh Welfare
g	=	Gram
GIT	=	Gastro intestinal tract
L	=	Liter
M.S.	=	Master of science

# LIST OF ACRONYMS AND ABBREVIATION (CONT'D)

ABBREVIATION		FULL MEANING
Ml	=	milliliter
Mg	=	milligram
NTFS	=	National Taskforce on Food Adulteration
SAU	=	Sher –E-Bangla Agricultural University
SS	=	Salmonella-Shigella
TCC	=	Total Coliform Count
TSC	=	Total Salmonella Count
USDA	=	The United States Department of Agriculture
WHO	=	World Health Organization

SYMBOLS		FULL MEANING
&	=	And
@	=	At the rate of
°C	=	Degree Celsius
۰F	=	Degree Fahrenheit
/	=	Per
<	=	Less than
lbs.	=	pounds
Tk	=	Taka
h	=	Hour
\$	=	Dollar
~	=	Approximately
%	=	Percentage
:	=	Ratio

## LIST OF SYMBOLS

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## HYGIENE PRACTICES OF TABLE EGGS AT DIFFERENT WET MARKET IN SHER-E-BANGLA NAGAR AREA OF DHAKA NORTH CITY CORPORATION: A PUBLIC HEALTH CONCERN

#### ABSTRACT

The study was carried out at different wet market of table eggs in Sher-e-Bangla Nagar area of Dhaka North City Corporation focusing on the hygienic practice of table egg vendors. It was done by survey and lab test of microbiological analysis of eggshell surface. A questionnaire was used that included the knowledge about food hygiene, egg handling, collection, transportation, storage, disposal procedure etc. Survey findings revealed that, most of the vendors of the selected area completed primary education level (52%), whereas only 6% completed Higher Secondary Certificate (HSC), 34% completed Secondary School Certificate (SSC), and the rest 8% were illiterate. Although many of them were literate, their knowledge about food hygiene was found to be very poor. The results showed that, only 4% vendors wash their hands before and after handling of egg where 96% vendors didn't wash hands before and after handling of eggs. It also revealed that, only 6% vendors dumped the broken eggs into drain whereas most of the vendors (94%) sold the broken eggs to customer, hotel and restaurant. So, there was a great chance of spreading the diseases if the eggs were contaminated. After completing the survey, laboratory test was conducted to find out pathogen presence in eggshell surface. Different egg samples were collected from different wet market of Sher-e-Bangla Nagar area and analyzed for Total Salmonella Count (TSC) and Total Coliform Count (TCC) in egg shell surface. These microbial test was performed to enumerate bacterial load and to determine the contamination and unhygienic conditions of egg handling. A total of 50 eggs (25 dirty & 25 clean eggs) were subjected to identify the presence of bacteria on eggshell surface. The highest value of TSC, TCC of both dirty eggs and clean eggs of eggshell were log 6.3, log 6.34, log 5.00, and log 5.11, respectively. The bacterial load was found highest in dirty eggs compared to clean eggs. Therefore, good hygienic practice is essential for safe handling of eggs. For this reason, appropriate action should be taken among the egg vendors as soon as possible by the government, particularly by the municipal authority.

#### **CHAPTER 1**

#### **INTRODUCTION**

#### 1.1 Background of the study

Food is a major cause of a significant number of diseases around the world. Bangladesh, a third world developing country in South Asia, is no exception. Consumption of unsafe food has been a major public health threat in Bangladesh for several decades. Adulterated foods have many deadly effects. The National Task Force on Food Adulteration (NTFS), established by GoB, finds that adulterated foods cause various food-borne diseases including diarrhea, malnutrition and other diseases every year, leading to the deaths of many people in Bangladesh. Children in particular are more vulnerable than adults as unsafe food is the leading cause of death in infants. The present study was therefore undertaken to identify the hygiene practices of vendors operating in various wet markets and to assess the nature of surface contamination and bacterial load of eggs (dirty and clean) in selected wet markets in the Sher-e-Bangla Nagar Area of Dhaka North City Corporation.

Eating dirty and unwashed eggs and egg products causes foodborne diseases such as salmonellosis, foodborne illnesses caused by *Salmonella enteritis's, Escherichia coli* etc. Eggs are contaminated at any stage of production like collection, transportation or marketing through vertical or horizontal transmission. In Bangladesh, egg contamination comes with significant healthcare costs. The lack of hygiene education of consumers and sellers is responsible for the problem of food safety. Therefore, good hygiene practices should be followed at every stage of egg production through to sale. It is therefore necessary to maintain good hygiene practices and to monitor the level of infection in the marketing cycle of eggs and egg products.

#### 1.2 State the problem

Today food safety is a serious public health concern around the world, including Bangladesh. Eggs and egg products play an important role in human health, especially in developing countries like Bangladesh. Eggs are fortified with proteins, minerals, fats and various vitamins such as vitamin  $B_{12}$ . According to the American Heart Association, the lutein found in the yolk also protects against the development of early heart disease. Recent studies show that dietary cholesterol, such as that found in eggs,

has very little effect on blood cholesterol levels. Healthy adults can eat an egg every day without increasing their risk of heart disease. The American Heart Association recommends that healthy adults consume no more than 300 milligrams of cholesterol per day. Eggs are a relatively low-calorie food and can be a great option for those on a diet. Because of its filling properties (the ability to keep you fuller longer), eating eggs for breakfast may promote a healthy weight and reduce the risk of obesity. However, in recent years, food contamination has become a serious global problem, and this problem also affects eggs. In fact, among food products, poultry and sea turtle eggs are among the most common sources of bacterial and fungal pathogens, as well as chemical contaminants such as synthetic organic compounds and heavy metals. While eggs are often considered excellent, other seabirds and turtles are actually among the most contaminated foods on the planet. According to the World Health Organization, approximately 1.5 billion cases and over 125,000 childhood deaths occur each year due to contaminated water and food. . According to Annor & Baiden (2011), in developing countries, an estimated 70% of diarrheal episodes are linked with the ingestion of contaminated foods. In 2017, WHO estimated that approximately 600 million people fall ill after eating contaminated food resulting in 33 million healthy years; foodborne diseases account for 40% diseases burden among under 5 years.

Food contamination is not only widespread in developing countries but also in developed countries of the world. In Bangladesh, small farms are the main source of eggs. As consumption of eggs has increased dramatically over the past two decades, they can be seen as the go-to choice for organized groups and NGOs. The shell of eggs are generally ineffective in preventing entry by microbes and some chemicals. Egg can be contaminated at both egg shell and egg contents by a variety of microbes with a wide range of pathogens such as *Campylobacter jejuni, Listeria monocytogenes, Escherichia coli, Yersinia enterocolitica and especially salmonella* (Ricke *et al.*, 2001; Board and Tranter, 1995). Contaminants include bacteria, mold and yeast. Contamination is more likely linked with cracked egg, dirty shells and storage in contaminated surroundings. It can be contaminated during formation and laying process (Abdullah, 2010). Elliott (1954) revealed that stored or aged eggs have more possibility to become infected than fresh eggs due to the degradation of natural defense mechanisms in egg over time. The eggshell contamination increasing the chances of egg contents contamination by penetration (Messens *et al.*, 2006). Bacterial contamination can happen at three main

parts of egg (egg yolk, albumen and shell membrane / egg shell) (Bahrouz, 2005). *Salmonella enteritis* is able to invade the cells of the follicles before ovulation and multiply themselves after 2 h of infection (Howard *et al.*, 2005). Eggs are considered to be a medium to low risk food for foodborne illness which can become contaminated with bacteria, like *Salmonella* and other enteric pathogens (Chousalkar *et al.*, 2010). A high wide variety of instances and outbreaks of salmonellosis are related with the consumption of eggs and egg products. The European Food Safety Authority (EFSA) and the European Centre for Disease Prevention and Control (ECDC) reported an overall European Union (EU) notification rate of 20.1 cases per 100,000 population, corresponding to 91,857 confirmed cases in 2018. (EFSA & ECDC, 2019)

In our country, small-scale laying farms are leading with weak biosecurity practices, in contrast to other large-scale commercial production systems that increase the risk of infection. FBD encompass various diseases that are responsible for morbidity and mortality worldwide. Egg-borne infectious diseases are a major public health concern worldwide and there are many outbreaks of foodborne illnesses, particularly those of a gastrointestinal nature, caused by the consumption of undercooked and contaminated eggs. Of the various foodborne pathogens associated with poultry products, gastrointestinal infections caused by egg-borne Salmonella other than typhoid, are the major concern in both developed and developing countries. The most common foodborne pathogens associated with food of animal origin are *Salmonella, Campylobacter, Listeria monocytogenes, Staphylococcus aureus and Escherichia coli* O157:H7 (Akbar and Anal, 2013a; Ghasemian, 2011; Akbar and Anal, 2011) Transmission of the disease from poultry to humans is suspected. The risk of egg-borne diseases is greatly increased by unsanitary egg production conditions and poor handling of eggs, including storage time and temperature.

#### 1.3 Significance of the study

The findings of this research would be great importance to vendors, researcher and the government in general. To vendors, it will provide knowledge on importance of hygiene practice and help them make correct decision on egg handling which reduces spreading infection to humans.

To researcher and government, it will help them to take a set of regulations and policy that should be implemented to ensure good hygiene practice by the vendors, working in different wet markets. If all the necessary precautions are not taken during the poultry production, marketing and processing chains in that case poultry meat and eggs can be contaminated by infectious agents that are harmful to humans.

So, this study holds a great importance to understand the present risks of egg borne diseases on human health and so that necessary measures will be taken and implemented in order to debilitating effect of foodborne disease by creating public awareness, increased monitoring, improving knowledge in vendors and consumers through good hygiene practice.

#### **1.4 Objectives**

Considering the above facts, the objectives of this research are-

- □ To find out the vendors' perception on table eggs cleanliness
- □ To assess the processing and preservation method of table eggs before sale
- □ To determine the pathogenic bacteria from table eggs available at different market in Sher-e-Bangla Nagar Area of Dhaka North City Corporation.

#### **CHAPTER 2**

#### LITERATURE REVIEW

This chapter gives a review of similar surveys that are related with the aspect of hygiene practice among egg vendors. It includes the present status of hygiene practices at different wet markets in Bangladesh & worldwide. It starts with first reviewing of experts thoughts regarding the study topic and it also looks at factual findings and conclusions that arise as a result of these works. Some areas of review include: concept of hygiene practice, demand of eggs & it demand, wet market condition, scenario of foodborne illness, role of eggs in foodborne disease, public health significance, and importance of hygiene practice.

#### 2.1 Concept of hygiene practice

Hygiene practices are the set of requirements to inhibit contamination of food in order to provide safe food to the consumers. Hygiene is an old idea associated with medicine, as well as to self and professional care practices related to most aspects of living. In medicine and in the home (domestic) and everyday life settings, hygiene practices are used as hindering measures to reduce the incidence and spreading of disease (Monney *et al.*, 2013).

Ababio *et al.* (2012), found that food is said to be hygienic when it is free of a hazardous substance that could be harmful to human or animal health. According to Fosket and Ceserani (2007), hygiene is the science and practice of maintaining health and preventing diseases in every catering establishment that provides food. Suffice it to mention that conscious effort must always be put in place to protect people as well as the environment from bacteria in order to eliminate contamination of the food that people eat (WHO, 2010)

Annor *et al.* (2011) in their study, they revealed that good hygiene practices have been documented to prevent several food-borne diseases when practiced. It is broadly acclaimed that deliberate or accidental contamination of food due to inappropriate handling of food might endanger the lives of consumers.

Odonkor *et al.* (2014), conducted a study of hygiene practice and concluded that several hygiene practices such as poor personal and environmental hygiene, inadequate storage of food and drinks improper preparation and cooking are known to cooperate the safety of food.

#### 2.2 Concept of table eggs nutritive value and demand

Mohammad *et al.* (2010), suggested that Poultry and poultry products become as a cheap source of animal protein in terms of meat and eggs. Poultry products especially the eggs and egg products plays vital nutritive role in human health specifically for developing country like Bangladesh (Vaclavik and Christian, 2014).

Table eggs are the best and easy source of food, containing quality protein, essential amino acids, essential vitamins and minerals needed for a good health (MAFF, 2009). Asia is the largest egg producing region with 65% global outputs (Ernst, 2009).

According to Song and Kerver (2000), Eggs contain a variety of important vitamins, minerals and trace elements. Layman *et al.* (2009), suggested that Eggs provide rich natural source of high quality protein and a number of vital nutrients including vitamins A, B<sub>12</sub>, B<sub>5</sub>, D, folate, choline, phosphorus, selenium and good amounts of vitamins E, K, B<sub>6</sub>, calcium and zinc. Eggs provide the richest mix of essential amino acids which is important for children, adolescents.

Bufano *et al.* (2000), suggested that eggs are considered one of the most delicious and popular foods all over the world; they can be prepared in many different ways to suit everyone's taste. Eggs and egg products enter in a wide variety of foods including custard, mayonnaise, egg salad and all types of bakery products. Also there are another uses of eggs as soil fertilization, culture media, artificial insemination, cosmetics, shampoo and adhesives. Eggs are super foods which provide unique well balanced nutrients for human of all ages. They contain large amounts of protein, amino acids, vitamins and minerals, low caloric value and easily digestible make them valuable choice in many therapeutic based diets for adults. Eggs can make a significant contribution to a healthy diet. A medium-sized egg provides 78 kcal, yet contains 6.5 g protein. The fat content is 5.8g of which 2.3 g is mono unsaturated fat.

Eggs have been identified to represent the lowest-cost animal source for proteins, vitamin A, iron, vitamin B<sub>12</sub>, riboflavin, choline, and the second lowest-cost source for zinc and calcium (Drewnowski, 2010). In addition to providing well-balanced nutrients for infants and adults, egg contains a myriad of biologically active components. The ten leading egg-producing countries are (in order): China, the United States, Indonesia, India, Mexico, Brazil, Russia, Japan, Turkey, and Pakistan, which together contributed approximately 75% of total global egg production ( $\sim$ 1,652 billion eggs, 99 million

tons) in 2019 (FAO, 2020). On a per capita basis, Americans consume about 234 eggs per year (American egg board, 2000).

According to USDA, it was at about 60 billion eggs in 1984 and at 67.3 billion eggs in 1998; Generally, about 70 percent of the edible shell eggs produced are sold as table eggs, while the remainder are processed into liquid, frozen, or dried pasteurized egg products. The majority of egg products are destined for institutional use or further processing into foods such as cake mixes, pasta, ice cream, mayonnaise, and bakery goods.

According to FAO, minimum of 104 eggs should be eaten by each person annually. The department of livestock services said that the per capita consumption of eggs was 121.18 in 2021 and 104.23 in 2020. Bangladesh has crossed the limit of minimum egg consumption rate set by FAO.

#### 2.3 Scenario of hygienic condition in different wet market

In developing countries, most of the people still depend on wet markets to purchase their daily food supplies. Soares *et al.* (2012), pointed that wet markets thus play a vital role in supplying fresh meat, eggs, vegetables, and fruits to the local population. Unfortunately, in some cases, wet markets have also had negative health implications. Wet markets are very common in South Asian countries including Bangladesh.

Nidaullah *et al.* (2017), in their study indicated that the traditional wet markets are commonly dirty, chaotic, unhygienic, and floors are constantly sprayed with water for washing and to conserve the humidity. Microorganisms on wet surfaces at wet markets can accumulate, convert into micro colonies and produce the biological film.

According to Chmielewski & Frank (2003), microorganisms within biofilms are protected from sanitizers increasing the likelihood of survival and subsequent contamination of food. This increases the risk of reduced shelf life of food and disease transmission.

Siddiky *et a*l. (2022), suggested that wet markets can be considered one of the hotspots for the spread and contamination of foodborne pathogens due to poor sanitary and hygienic measures and practices.

Nagan *et al.* (2020), stated in his studies that these informal marketplaces are more sustainable, as they reduce transport and limit waste and the amount of packaging. It is

surely important to consider that, in each form of food product trade, hygienic practices must be guaranteed and implemented to ensure food safety and public health.

#### 2.4 Scenario of causes and result of food borne disease

Foodborne illness is a major public health problem and the main cause of diarrheal diseases affecting all developed and developing countries (Akbar and Anal, 2014; Akbar and Anal, 2013b).

Foods most likely to cause human illness are animal products such as red meat, poultry and eggs, seafood, and dairy products. Foodborne pathogens are major concerns of food safety Foodborne illnesses from six bacterial pathogens are estimated to account for \$2.9-\$6.7 billion in human illness costs in the United States each year. The six bacterial pathogens studied in this report all found in animal products are *Salmonella, Campylobacter jejuni, Escherichia coli O157:H7, Listeria monocytogenes, Staphylococcus aureus, and Clostridium perfringens.* They are responsible for approximately 76 million cases of foodborne illness, 325,000 hospitalizations and 5,000 deaths in the U.S.A annually (Buzby *et al.*, 1996).

According to World Health Organization an estimated 1.5 billion cases and over 125, 000 deaths occur in children every year as a result of water and food contaminations. In developing countries, an estimated 70% of diarrheal episodes are linked with the ingestion of contaminated foods. (Mukhopadhyay *et al.*, 2012 & Annor *et al.*, 2011).

Worldwide, there are nearly 1.7 billion cases of diarrheal disease every year. Diarrheal disease is the second leading cause of death in children under 5 years old. Every year about 760,000 children under 5 years old die due to diarrheal diseases (Chowdhury *et al.*, 2015)

Usfer *et al.* (2010), in their study indicated that unsafe drinking water and improper food handling practices lead to diarrhea which is the second leading cause of child mortality worldwide. Each year more than 1.5 million children under the age of 5 die of acute diarrhea, which translates in 2000-2003 to 18% of deaths of children under the age of five. Schlundt *et al.* (2004), pointed out that food borne diseases are increasing in both developed and developing countries. Diarrheal diseases are mostly caused by food borne microbial pathogens, which are a leading cause of illness and deaths in the developing countries, killing an estimated 1.9 million people annually at the global level.

# 2.5 Present scenario in contamination rate of bacteria (*Salmonella* and *E.coli*) in Bangladesh

Foodborne pathogens are often associated with outbreaks and can affect a wide variety of nearby people. Many attacks are attributed to different *Salmonella* serovars each year, demonstrating the prevalence of enteritis cause by *Salmonella enterica* serovar *typhimurium* (Kariuki *et a*l., 2006).

Monzur and Ahmed (2011), reported that Bangladesh is one of the developing country in the world, but rich in poultry industry as other country like India and Pakistan. The Centers for Disease Control and Prevention (CDC) reported in 2013 that in recent year's public health problems associated with salmonellosis were of poultry origin. Several studies were done regarding prevalence of *Salmonella* from egg collected from poultry and market. *Salmonella* was prevalent in a wide range (8%-12%) from egg in Dhaka city.

The contamination rate of different serotype of *Salmonella* in 4.47% of market eggs and 6.66% of poultry eggs. Of them, most of the identified *Salmonella* serotype was *S. Enteritis*. (Fardows & Shamsuzzaman, 2015). Prevalence rate of *Salmonella* is 83% in poultry eggs in different market of Savar area (Mahmud *et al.*, 2015). Contamination mainly by moist environment of market and poultry house, poultry feeds, unhygienic practice of farm handlers and surrounding environment. (Safaei *et al.*, 2011). The *Salmonella* contamination rate from poultry feeds, where 71.43% *Salmonella* was detected from poultry feeds from different market in Savar area. (Chowdhury *et al.*, 2011)

When eggs are exposed to a contaminated environment and bacteria breach the eggshell, horizontal transmission occurs. *Staphylococcus aureus, Salmonella* spp., *Streptococcus spp., Escherichia coli, Bacillus spp.,* and *Listeria monocytogenes* were among the bacteria detected on the eggshell (Mahdavi *et al.,* 2013).

*E. coli* contaminations are more likely with a cracked eggs, dirty shells and storage in contaminated surroundings. Contaminating egg shells increased the changes of egg contents when the shells are broken (Neira *et al.*, 2017).

The findings of Islam *et al.* (2008), who reported that 34.64% chicken eggs were contaminated with *E. coli* in Dhaka city of Bangladesh. Hossain *et al.* (2021), in their study found that the overall prevalence of *E. coli* was 38.89% in chicken eggs in

Rajshahi district of Bangladesh.

#### 2.6 Public health significance of salmonellosis

Jong *et al.* (2006), pointed that salmonellosis is an important global public health problem causing substantial morbidity and thus also has a significant economic impact. Although most infections cause mild to moderate self-limited disease, serious infections leading to deaths do occur. Voetsch *et al.* (2004), also stated in his article that Salmonellosis is one of the most important food-borne disease and causes substantial medical and economic burdens worldwide

Several foods have been linked to cases and outbreaks of salmonellosis (CDC, 2020b; EFSA & ECDC, 2019; Meinen *et al.*, 2019). However, in 2018, 45.6% of the reported salmonellosis outbreaks in the EU were associated with the consumption of "eggs and egg products" (EFSA & ECDC, 2019). In addition to its impact on health, Salmonella and its associated illnesses have become an economic burden to society. The average annual cost of medical care and loss of productivity due to Salmonella infection in the United States ranges from \$0.5 to \$4.4 billion (Scharff *et al.*, 2012).

In spite of the improvement in hygiene, food processing, education of food handlers and information to the consumers, foodborne diseases still dominate as the most important public health problem in most countries (Mather *et al.*, 2013) .This has significant implication in the developing countries like Bangladesh where poultry industry is the fastest growing segments (Hoelzer *et al.*, 2011).

In a meta-analysis conducted by Domingues *et al.* (2012), consumption of undercooked or raw eggs and poultry was found as a risk factor for sporadic cases of salmonellosis. Olsen *et al.* (2013), explained in their study that salmonella is mostly transmitted to humans, through contaminated food and water. In hospitals, person to person transmission may also happen. Cross contamination can occur in farm houses as well as during handling of poultry products. *Salmonella* can also leads to severe condition like sepsis and death especially in infants and immunocompromised adults (Tessari *et al.*, 2015).

Andino *et al.* (2015), in their study, pointed that Other than gastroenteritis, *Salmonella* may also cause extra intestinal infection like meningitis, osteomyelitis, arthritis, pneumonia, cholecystitis, peritonitis, pyelonephritis, endocarditis, pericarditis, vasculitis and chronic condition like aseptic arthritis and Reiter's syndrome. The

predominant servers of *Salmonella*, having public health importance are mainly *S. enteritis* and *S.Typhimurium*. Recent concern in public health point of views antibiotic resistant serotypes (Ferdous *et al.*, 2013).

The WHO observed an alarming rate increment of resistant *Salmonella* strains due to the abusive use of antibiotics in poultry farm. The horizontal transmission of virulence genes in multidrug resistant *Salmonella* strains can increase virulence and invasiveness and it cause high mortality rates (Han *et al.*, 2011).

#### 2.7 Public health significance of disease producing coliform bacteria

Food borne diseases are a growing public health problem all over the world which cause an estimated 48 million illnesses and 3,000 deaths each year in the United States (Scallan *et al.*, 2011). In developed countries, up to 30% of the population suffer from food borne diseases each year, whereas in developing countries up to 2 million deaths are estimated per year (WHO, 2007a and 2007b).

*E. coli* is one of the predominant food borne bacterial pathogen. Majority of the *E. coli* are non-pathogenic but few of them are enormously pathogenic causing watery and bloody diarrohea e.g., *E. coli* 0157:H7 that's associated with lifestyles threatening disease hemorrhagic colitis, hemolytic uremic syndrome and thrombotic thrombocytopenic purpura. *E. coli* 0157:H7 infection can lead to hospitalization because of hemorrhagic colitis and, in severe cases, hemolytic uremic syndrome. (Nguyen *et al.*, 2012).

*E. coli* is most common species of facultative anaerobe found in the GTI of both man and animals and the most commonly encountered pathogen in the Enterobacteriaceae family, therefore the presence of such organism in foods is the indication of fecal contamination (Mohamed *et al.*, 2014).

*E. coli* bacteria are enteropathogenic and/or toxic microorganisms harmful to human health. Enteropathogenic bacteria are gram-negative bacteria, pathogenic in nature, that attack the human digestive system (Pratama KA *et al.*, 2020). Certain strains of *E. coli* bacteria can cause gastroenteritis in humans, where gastroenteritis is a disease of the digestive system such as vomiting and diarrhea caused by infection, which are bacteria that cause digestive disorders such as nausea, vomiting, and diarrhea (Ruth Melliawati, 2014). Salauddin (2015), stated that isolated *E. coli* in all the samples of broiler and found some isolates of E. coli were multi drug resistant. Islam *et al.* (2010), characterize

shiga-toxin producing *Escherichia coli* in raw meat, raw milk in Bangladesh. Food borne diseases including colibacillosis are a significant and widespread global public health threat reported in Bangladesh (Samad, 2011; Jakaria *et al.*, 2011).

#### 2.8 Scenario of hygiene knowledge and practices

It has been argued that a good knowledge of food hygiene will lead to proper food handling and proper preparation practices by food suppliers. On the other hand, poor food hygiene knowledge can lead to improper handling of food ingredients. While preparing food, catering workers have direct contact with food and some wear jewelry. Others may touch their bodies while cooking, causing food contamination and food poisoning .Globalization of food trade has focused its attention on strengthening measures taken to ensure quality and safety, especially on finished foods, and different countries have developed specific regulations to ensure food safety and hygiene (Ojinnaka, *et al.*, 2011).

Food poisoning occurs because contaminated food is consumed. Foodborne illness is considered a major public health problem with a significant impact on economic losses. Millions of people get sick each year, and hundreds to thousands die after eating contaminated food due to poor handling and safety practices by food retailers. Food poisoning and food poisoning from parasites and pathogenic bacteria can occur as a result of improper food and food handling, food preparation and storage, and contamination can occur. When an infected person is involved in food preparation, the likelihood of food contaminated foods is the main cause of more than half of diarrheal diseases most communities in developing countries.

Osagbemi *et al.* (2010), in their study revealed that the number of reported cases of food poisoning has been increasing in recent years and many of the outbreaks can be traced to contamination caused by poor food hygiene among people.

Food and personal hygiene are known to prevent several food-borne diseases when practiced. It is broadly acclaimed that deliberate or accidental contamination of food due to improper handling of food might endanger the lives of consumers. Several hygiene practices such as poor personal and environmental hygiene, inadequate storage of food and drinks, improper preparation and cooking are known to compromise the safety of food (Annor and Odonkor, 2011).

World Health Organization (2007), reported that food safety is an essential public health issue for all countries. Food borne diseases due to microbial pathogens, bio toxins and chemical contaminants in food represent serious threats to the health of thousands of millions of people.

Lin *et al.* (2003), stated that the Centres for Disease Control and Prevention (CDC) reported that hand washing is one of the most important hygienic procedures in preventing the transmission of infectious disease. It is the first line of defense for infectious diseases, including respiratory infection and gastrointestinal disorders among others.

An important way to prevent food contamination is to maintain a high standard of personal hygiene and cleanliness. Mishandling of food and disregard of hygienic measures on the part of food handlers may enable pathogens to come into contact with food and in some cases, to survive and multiply in sufficient numbers to cause illnesses in the consumers (Zain and Naing, 2002).

#### 2.9 Knowledge about public health hygiene in Bangladesh

According to Vriesekoop *et a*l. (2010), a principal contamination risk factor of public health concern linked to food handling is the concurrent handling of food and money because of the presence of certain pathogens e.g. *E. coli, S. aureus* and *Salmonella* spp. on banknotes or currencies. The research also showed that fingernail hygiene practices differ from one geographical area to the other the tendency of food vendors to properly wash their hands also depends on access to improved water sources.

Ali (2013), reported that most of the food processor or shop keepers are aware about the public food hygiene but they have the tendency to violate the roles due to regulatory reasons.

Various socio-economic, cultural, and regulatory reasons may be liable for these food safety problems in Bangladesh, such as lack of consumer education (Huda *et al.*, 2009), product price (Hossain *et al.*, 2008), multiplicity of laws, lack of coordination, lack of adequate punishments, judicial restraint (Andaleeb and Ali, 2009). Therefore, the strong commitment and regulations of government may control these problems. Ultimately deficiencies in hygiene practices may be attributable to inadequate food regulation enforcement by the local authorities.

#### 2.10 Scenario of hygienic food condition in Bangladesh

Unhygienic practices in food handling become a common phenomenon in the Bangladesh food industry. In another report, it was revealed that in Bangladesh at least 501 people visits hospital every day for diarrheal diseases that are related to food safety (Ali, 2013). This long diarrheal disease problem, related to food safety, persists and it has been a significant cause of malnutrition in Bangladesh for the last couple of decades (DGHS, 2012).

Huda *et al.* (2009), pointed that there are some reasons such as lack of consumer and food processor education is liable for food safety problems in Bangladesh.

In Bangladesh, consumption of unsafe food is a serious problem to public health. A survey conducted by the Institute of Nutrition and Food Science, Dhaka University, in early 1980s had revealed that 60 per cent of the people of Bangladesh are suffered in malnutrition due to intake of inadequate diets and adulterated food, eggs etc.

The report of the Directorate General of Health Services (DGHS) mirrors the magnitude of the diarrheal diseases and confirms that this health problem is caused by mainly unsafe foodstuffs. The DGHS report suggests, from 2003 to 2009 17,999,284 people were attacked by diarrhea and among them 4,674 people died, 16 which signifies that in average at least 3,850 people die for diarrhea each year.

### **CHAPTER 3**

### **MATERIALS AND METHOD**

The study was performed to find out the hygiene practice of table eggs at different wet market in Sher-e- Bangla Nagar area of North City Corporation and microbiological analysis to find out bacteria (*Salmonella* and *E.coli*) present on the egg shell surface.

#### 3.1 Research Design

A survey was done about the hygiene condition concerning egg cleaning procedure, preservation & processing techniques, disposal procedure knowledge was performed between June2021 to July 2021.

Laboratory analysis of table egg was done. There are two assessment system were followed during the experiment. These are

- To perform an experiment to find out the hygiene practice among vendors at different wet market in Sher-e- Bangla Nagar area.
- Assessment of microbiological analysis of egg shell and bacterial count in egg shell surface by lab test.

#### 3.1.1 Experimental location

The experiment was performed at different wet market in Sher-e- Bangla Nagar area of North City Corporation.

#### 3.1.2 Sampling procedure

The estimated number of egg vendors that operate at different wet markets in Dhaka city is 50. A sample size of 50 vendors were selected from different market randomly was interviewed. The researcher targeted those people who were regularly sell different types of egg such as duck, hen, quail etc. This population was chosen because they are considered as high risk populations, which are at higher risk of suffering from different disease though handling eggs if the hygiene practices are not followed by egg vendors.

#### 3.1.3 Questionnaire

Considering the objective, the appropriate research instrument was a questionnaire to determine the effects of hygienic practices of vendors on food safety. Collis and Hussey (2003), contend that the questionnaire is made to come out with the doing, thinking or feeling of a selected group of individuals. For this reason, a structured questionnaire

made up of 18 questions was administrated to the vendors. The questionnaire was outlined by research supervisor. Respondents were interviewed to determine their general knowledge about food hygiene, disposal procedure of broken, rotten and dirty eggs. Selling pattern about table eggs such collection of eggs, transportation, storage time, storage condition, packing system were also investigated.

#### 3.1.4 Sampling unit

The sampling unit or respondent was a vendor of wet market in Sher-e- Bangla Nagar area of Dhaka North City Corporation

#### 3.1.5 Sample size

The required sample size was determined by the level of confidence and precision to be followed. The advantage of this approach is that the statistical significance of the sample does not depend on the size of the sample relative to the population studied. Rather, it is the required level of probability (confidence level), the required level of accuracy, and the population variability that matters. The required sample size was estimated using the following formula (S.K. Lwanga, 1991):

$$n = \frac{Z^2 P (1-P) N}{Z^2 (N-1) + Z^2 P (1-P)}$$

n = Required number of sample size =50

Z= Confidence level 95% = 1.96

P=0.5

E= Design effect (0.131)

N= Number of wet markets in sampled area = 471

#### 3.1.6 Sample Distribution

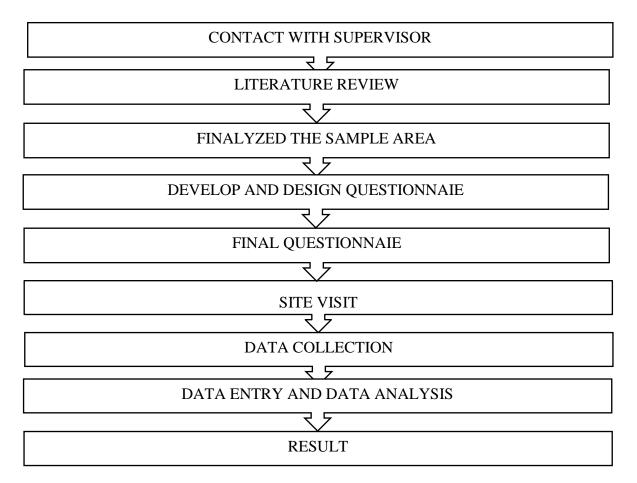
The experiment randomly selected the vendors of different wet market in Sher-e-Bangla Nagar area.

#### 3.1.7 Data Collection Instrument

Questionnaire was the survey instrument.

**D** Questionnaire for table egg vendors (Attached in appendix)

#### 3.1.8 Survey Implementation



#### Data collection-

- General information about vendors
- Vendors concern about hygiene practice
- Information about selling pattern of table eggs
- > Information about the disposal process of broken and rotten eggs

#### Data entry

Data entry was conducted by with the supervision of research Supervisor.

#### Giving gift at the end of questionnaire

A small gift was given after the end of questionnaire.

#### 3.1.9 Data Processing and Presentation

Data was imported to the Microsoft Access and Microsoft Office Excel. A simple graphical design and tabular technique will be presented in the study to classify the data into meaningful categories with estimates of different parameters.

#### 3.2 Microbiological analysis

#### 3.2.1 Sample collection

Eggs (brown eggs) were collected randomly from different wet market of Sher-e-Bangla Nagar Area over a period of 1 month. A total of 50 eggs were analyzed to detect the presence of *Salmonella* spp. and *Escherichia coli* on eggshell surface. The representative Eggs from different store for the study were collected aseptically using sterile polythene bags and transported to nutrition laboratory of Sher-e-Bangla University for analysis.

#### 3.2.2 Sample analysis

The collected sample are categorized into several regions and then analyzed for microbial detection. The microbiological analysis was carried out in the Animal Nutrition Laboratory in the faculty of Animal Science and Veterinary Medicine at Shere-Bangla Agricultural University, Dhaka-1207. The laboratory had enough facilities for the determination of microbiological analysis of egg sample.

# Table 1. No. of egg samples collected from selected areas of Sher-e-Bangla NagarArea

Serial No.	Place	No. of Collected Samples
1.	Agargaon Market	10
2.	Taltola Market	10
3.	Krishi Market	10
4.	SAU Market	10
5.	Bihari Kamp Market	10
Total		50

#### **3.2.3 Media for bacteriological study**

Bacteriological analysis were performed to determine the number of salmonella and E.coli present on eggshell surface by using *Salmonella-Shigella* Agar (SS Agar) and Eoisin methylene blue agar (EMB).

#### Salmonella-Shigella Agar (SS Agar)

*Salmonella-Shigella* Agar (SS Agar) agar mainly used as a selective and differential medium for identification of salmonella sand some *shigella* organisms. This SS agar media was made according to manufacturer company instructions (Hi media, India).

## Eosin Methylene Blue Agar (EMB Agar)

EMB agar was used to identify the *E. coli* organism present in the selected sample. EMB agar media was prepared according to the instruction of Manufacturer Company (Hi media, India)

## 3.2.4 Total salmonella count

The suitable dilutions of sample are blended with medium, whilst incubated at appropriate temperature that can assist the growth of microorganism. The overall *salmonella* count gives the estimate of number of salmonella found in egg sample. This approach has been set up via the use of conventional temperature and easy way of life medium. The bacterial colonies that develops on Petri dish improved by way of dilution component to symbolize the quantity of microorganisms present in sample tested. The test was also performed to determine the contamination and unhygienic condition of egg handling.

## Apparatus and reagents:

- □ Salmonella-Shigella Agar (SS Agar )
- □ Phosphate buffer solution (PBS)
- Petri dishes
- □ Incubator
- □ Autoclave
- □ Sprit lamp
- □ Colony counter
- **D** Egg sample
- □ Micro pipette
- □ Micro pipette tips
- **D** Eppendorf tube
- □ Electronic balance
- **O**ven
- **D** Distilled water
- □ Measuring cylinder

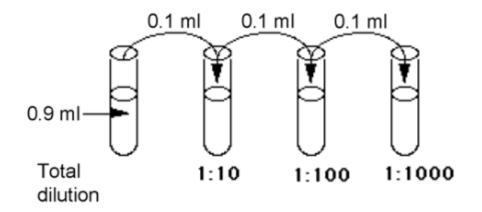
## **Procedure**:

#### **Step 1: Preparation of Samples**

- To detect *salmonella* from egg shell, a swabbing techniques were used for making inoculums of eggshell surface, a sterile cotton swabs wetted in PBS solution was used for surface swabbing and it was immersed into the eppendorf tube containing 1ml PBS solution.
- Mark all the petri dish with sample number, dilution and other desired information before making dilutions.

#### **Step 2: Preparation of Dilution Blanks**

- Dilution blank was prepared with PBS solution. To find out the microbial contamination of egg shell, sample collected from egg shell was mixed with 1ml of PBS solution in an eppendorf tube and mixed well.
- This made the individual sample. Then 0.1 ml of this sample was transferred to 0.9 ml of PBS solution to make 1:10 dilution. This was 10<sup>1</sup> dilution factor. Now 0.1 ml from the 1:10 ration in the first tube was transferred to second tube which contain 0.9 ml of PBS solution. The dilution factor was 10<sup>2</sup>.



Finally 1:1000, 1:10000, 1:10000 was made in the same way and the dilution factor was 10<sup>3</sup>, 10<sup>4</sup> and 10<sup>5</sup> respectively.

Ingredients	Gms/litre
Lactose	10.0g
Bile salts	8.5 g
Sodium citrate	8.5g
Sodium thiosulfate	8.5 g
Beef extract	5.0g
Casein peptone	2.5g
Meat peptone	2.5g
Ferric citrate	1.0
Neutral red	25.0mg
Brilliant green	0.33 mg
Agar	13.5g
Distilled water	1000ml

Step 3: Preparation of SS Agar: Composition of Salmonella-Shigella Agar (SS Agar)

- □ About 63 gm agar was suspend in 1000 ml of distilled water and was mixed thoroughly.
- □ Then the media was heat with frequent agitation and was boiled for one minute.
- $\Box$  There was no need to autoclave the media.
- ☐ Agar could be store in refrigerator for further use. A prepared culture media can be kept for at least a week in refrigeration.

#### **Step 4: Pouring the agar and sample**

- □ All the petri dish was sterilized by autoclaving at 15 lbs. pressure (121°C) for 15 minutes.
- Poured about 10-15ml of melted agar in each of petri plates containing the sample. Shake 10<sup>1</sup> dilution and delivered 0.1 ml to one Petri dish and do the same thing next all petri dishes.
- □ The agar was mixed with the sample by rotating and titling the dish. All petri dishes was allowed for some time for solidification.
- Separate sterile micropipette tips was used for transfers from each different dilution Page | 21

#### **Step 5: Incubation**

- □ The petri dishes were placed in inverted position and place them in the incubator at 37°C for 24h.
- □ Counted the plates after the incubation period. Record the dilutions used and number of colonies counted on each plate. If it is impossible to count at once, after the required incubation store the plates at 0 to 4.4°C for not more than 24 h.

#### **Step 6: Counting the bacteria**

- At the end of the incubation period, the petri plates containing between 30 and 300 colonies was selected.
- Plates with more than 300 colonies cannot be counted and are designated too many to count. Plates with fewer than 30 colonies are designated too few to count. Counted the colonies on each plate. A colony counter should be used.
- Calculating the number of bacteria (CFU) per milliliter or gram of sample is the number of colonies multiplied by the dilution factor divided by the amount of specimen added to the agar media.

#### Calculation

CFU/mL = (no. of colonies × dilution factor)/ volume of culture plate

#### **Counting bacteria**

After the completion of incubation, colonies are counted with the help of colony counter. The minimum size of bacteria was 0.5 mm.

#### Interpretation of the results

- Salmonella will not ferment lactose, but produce hydrogen sulfide (H<sub>2</sub>S) gas. The resulting bacterial colonies will appear colorless with black centers.
- Shigella do not ferment lactose or produce hydrogen sulfide gas, so the resulting colonies will be colorless.
- Coliform bacteria such as *E. coli* will ferment the lactose in the media, resulting in bacterial growth with a pink color. They do not produce any hydrogen sulfide.
- Enterobacter and Klebsiella appears larger than E. coli, mucoid, pale, opaque cream to pink.

## **3.2.5 Total coliform count**

Coliforms are the group of bacteria that are always present in the digestive tract of animals and humans and also found in their wastes. *Escherichia coli* (*E.coli*) is member of coliform bacteria that is commonly found in gut of humans and warm blooded animals. Most strains of *E. coli* produces toxin that can cause serious foodborne disease. Primary source of these outbreaks are raw milk, handling eggs contaminated with fecal material, undercooked meat products etc. EMB agar was used as a selective and differential medium for the isolation of coliform bacteria in foods, milk and clinical products. This agar contains eosin and methylene blue which inhibit the growth of many microorganisms. The gram negative bacteria (lactose fermenting) acidify the medium and under acidic condition, it produces a deep purple complex associated with green metallic sheen. The lactose non fermenters such as salmonella form a clear colonies that are easily detected and distinguished from coliforms.

#### Apparatus and reagents:

- Eosin Methylene Blue(EMB) Agar
- □ Phosphate buffer solution(PBS)
- Petri dishes
- □ Incubator
- □ Autoclave
- □ Sprit lamp
- **C**olony counter
- **D** Egg sample
- □ Micro pipette
- □ Micro pipette tips
- **D** Eppendorf tube
- □ Electronic balance
- Oven
- Distilled water
- □ Measuring cylinder

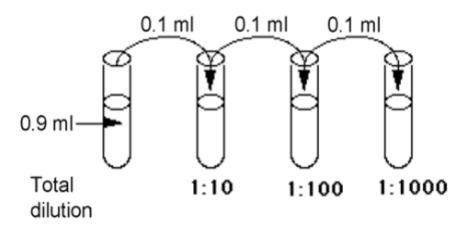
#### Procedure

#### **Step 1: Preparation of Samples**

- To detect salmonella from egg shell, a swabbing techniques were used .for making inoculums of eggshell surface, a sterile cotton swabs wetted in PBS solution was used for surface swabbing and it was immersed into the eppendorf tube containing 1ml PBS solution.
- Mark all the petri dish with sample number, dilution and other desired information before making dilutions.

#### **Step 2: Preparation of Dilution Blanks**

- Dilution blank was prepared with PBS solution. To find out the microbial contamination of eggshell, sample collected from egg shell was mixed with 1ml of PBS solution in eppendrof tube and mixed well.
- This made the individual sample. Then 0.1 ml of this sample was transferred to 0.9 ml of PBS solution to make 1:10 dilution. This was 10<sup>1</sup> dilution factor. Now 0.1 ml from the 1:10 ration in the first tube was transferred to second tube which contain 0.9 ml of PBS solution. The dilution factor was 10<sup>2</sup>.



□ Finally 1:1000, 1:10000, 1:10000 was made in the same way and the dilution factor was 10<sup>3</sup>, 10<sup>4</sup> and 10<sup>5</sup> respectively.

#### Step 3: Preparation of agar media

Composition

Ingredients	Grams/litre
Peptic digest of animal tissue	10.00
Dipotassium phosphate	2.00
Lactose	5.00
Sucrose	5.00
Eosin-Y	0.400
Methylene blue	0.065
Agar	13.5
Final pH (at 25 <sup>0</sup> C)	7.2±0.2

#### **Step 4: Pouring the agar and sample**

- □ All the petri dish was sterilized by autoclaving at 15 lbs. pressure (121°C) for 15 minutes.
- Poured about 10-15ml of melted agar in each of petri plates containing the sample. Shake 10<sup>1</sup> dilution and delivered 0.1 ml to one petri dish and do the same thing next all petri dishes.
- The agar was mixed with the sample by rotating and titling the dish. All Petri dishes was allowed for some time for solidification.
- Separate sterile micropipette tips was used for transfers from each different dilution.

#### **Step 5: Incubation**

- □ The petri dishes were placed in inverted position and place them in the incubator at 37°C for 24h.
- □ Counted the plates after the incubation period. Record the dilutions used and number of colonies counted on each plate. If it is impossible to count at once, after the required incubation store the plates at 0 to 4.4 °C for not more than 24 hours.

#### **Step 6: Counting the bacteria**

At the end of the incubation period, the petri plates containing between 30 and 300 colonies was selected. Plates with more than 300 colonies cannot be counted and Page | 25

are designated too many to count (TMTC). Plates with fewer than 30 colonies are designated too few to count (TFTC). Counted the colonies on each plate. A colony counter should be used.

Calculating the number of bacteria (CFU) per milliliter or gram of sample is the number of colonies multiplied by the dilution factor divided by the amount of specimen added to the agar media.

## Calculation

CFU/mL = (no. of colonies × dilution factor)/ volume of culture plate

## **Counting bacteria**

After the completion of incubation, colonies are counted with the help of colony counter. The minimum size of bacteria was 0.5 mm.

## Result interpretation on EMB agar.

- *Escherichia coli*: Blue-black bulls' eye; may have a green metallic sheen
- > Salmonella Typhimurium: Luxuriant growth; colorless colonies



SAU Market



Agargoan Market



Krishi Market

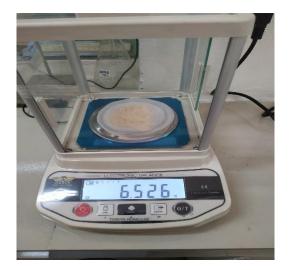


Taltola Market



Bihari Kamp Market

Plate 1. Some pictorial view of data collection





**Digital Balance** 

Micropipette



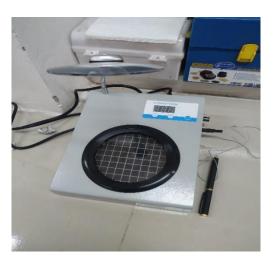
**Autoclave Machine** 



Magnetic Stirrer



Incubator



**Colony counter** 

# Plate 2. Equipment required for microbiological analysis

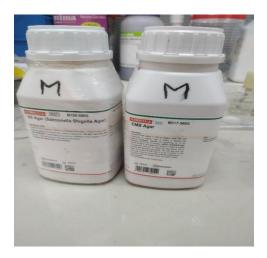


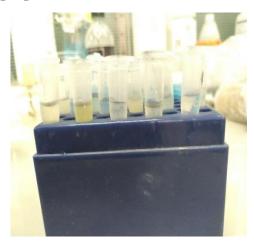


Plate 3. Agar media preparation



Plate 4. Autoclaving at 121°C by Autoclave machine





**Plate 5. Sample Preparation** 



Plate 6. Monitoring of research activities by the supervisor



Plate 7. Media preparation



Plate 9. Diluted sample were spreaded by glass spreader

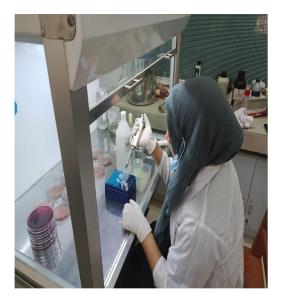


Plate 8. Dilution of sample

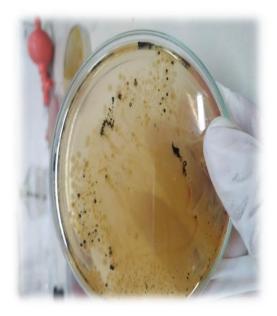


Plate 10. Incubation of media at 37<sup>o</sup>c in Incubator



Plate 11. Colony counting







Clean egg



Dirty egg

Plate 12. Bacterial colony found in SS agar



Dirty egg

Clean egg



In dirty egg, green metallic sheen colonies found in EMB Agar

Plate 13. Bacterial colonies found in EMB agar

# **CHAPTER 4**

# **RESULT AND DISCUSSION**

This study represent the result and analysis of data that are collected from different wet markets of Sher-e-Bangla Nagar area of Dhaka North City Corporation. Data were collected from 50 table egg vendors of different market of the selected area. We established a Questionnaire for table egg vendors and collected the sample from the surface of the eggshell for microbiological analysis. With the use of a table and graphs, the findings are displayed under the following heading.

## 4.1 Assessment of educational qualification and hygiene perception of vendors

Survey findings revealed that (figure 1) most of the vendors of the selected area had completed primary education level (52%), where only 6% completed HSC, 34% completed SSC, the rest 8% were illiterate.

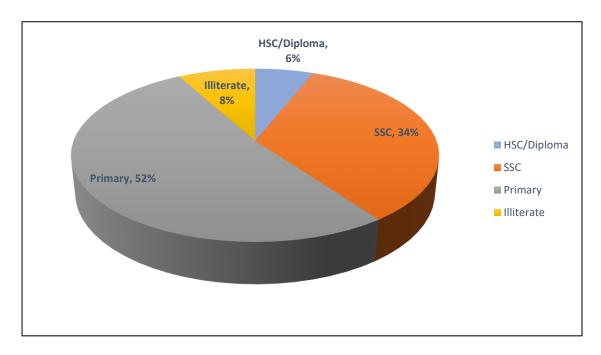


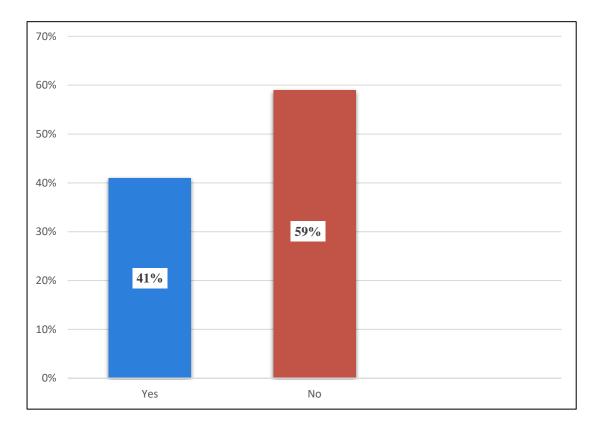
Figure 1. Education level of vendors

From this analysis, it was seen that the people involved in this sector, most of their education level is up to primary. This low level of education and low level knowledge of hygiene practices can contribute to poor hygiene practices by suppliers.

#### 4.1.1 Knowledge about the consumption of undercooked or raw eggs

Figure 2 indicates that among the vendors, 41% had the knowledge about consumption of raw and undercooked eggs have harmful effect to health and 59% had no knowledge about consumption of raw eggs have detrimental effect on health.

In the study of Sharif and Al-Malki (2010), found that half of the Saudi college students were not aware of the food poisoning risks associated with raw eggs consumption and believed that raw eggs are healthier than pasteurised eggs. Another study Odeyem*i et al.* (2019), reported that only half of the Jordanian students knew that consumption of risky foods like fried eggs with a runny or soft yolk, raw homemade cookie dough or cake batter can increase the risks of foodborne diseases.



#### Figure 2. Knowledge about the consumption of undercooked or raw eggs

According to Food and Drug Administration (FDA) recommends using pasteurized eggs. These eggs have been heated up enough to kill the *Salmonella* bacteria that was potentially inside. From the present findings it was revealed that those insufficient knowledge about health risk of raw egg consumption is probably a purpose of increasing foodborne illness.

# 4.1.2 Knowledge about disease spread through dirty eggs

Survey revealed that 68% (figure 3) had the knowledge about the dirty, broken egg can spread the disease. But only 4% (figure 4) vendors wash their hands before and after handling of eggs where 96% vendors didn't wash hands before and after handling of eggs.

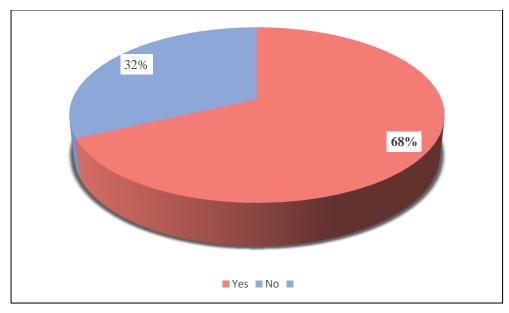


Figure 3. Knowledge about disease spread through dirty eggs

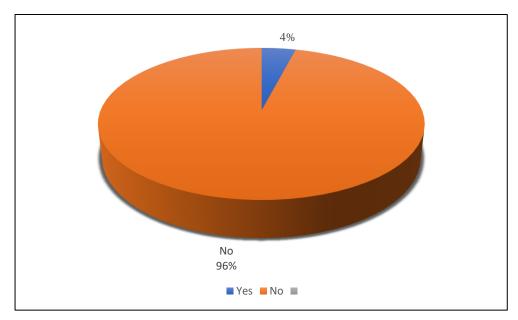


Figure 4. Wash hands after and before handling of eggs

*Lin et al.* (2003), stated that the Centers for Disease Control and Prevention (CDC) reported that hand washing is one of the most important hygienic procedures in

preventing the transmission of infectious disease. It is the first line of defense for infectious diseases, including respiratory infection and gastrointestinal disorders among others. Dasgupta (2005), in his study revealed that many food borne diseases and pathogenic microorganisms are spread by contaminated hands.

The present findings are contradictory with the findings of Whiley *et al.* (2017), a survey conducted in Australia, 38.7% of the respondents answered "always" to the question "How often would you wash your hands after handling eggs during food preparation. Another survey conducted in European countries, more than 50% of the respondents stated that they wash hands with soap after handling raw eggs, meat, poultry, or seafood (Koppel *et al.*, 2015).

## 4.1.3 Idea about cleaning eggs for selling

This result showed that that only 37% (figure 5) vendors clean their eggs before selling and 63% vendors do not clean their eggs before selling. If they clean the eggs, 69% (figure 6) vendors in follow the wet method and rest of them follow the dry method.

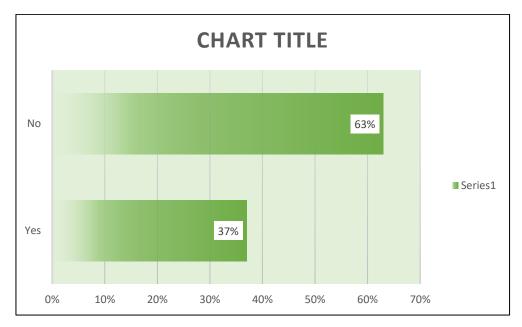


Figure 5. Cleaning eggs before selling

Hutchison *et al.* (2019), in their study found that eggs are normally washed with warm water and sanitized with industry recommended sanitizers before being delivered to the hatchery, but there may be variations in the effectiveness of this practice depending on operators. For commercial table eggs, washed eggs may be treated as higher risk

because washing can introduce contamination into an egg. From this survey, it was concluded that wet cleaning of eggs might be a cause of egg contamination.

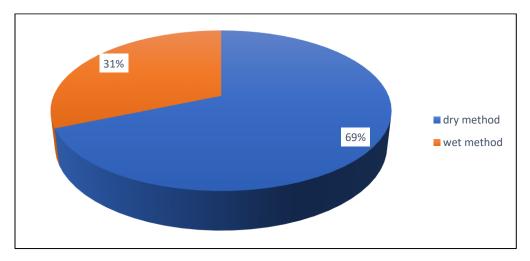


Figure 6. Types of cleaning method

The survey revealed that 43 % (figure 7) vendors keep the raw eggs close to other products such as vegetable. So, there had a great chance of spread the diseases if the eggs are contaminated. This result indicated that most of the vendors were apathetic about food safety.

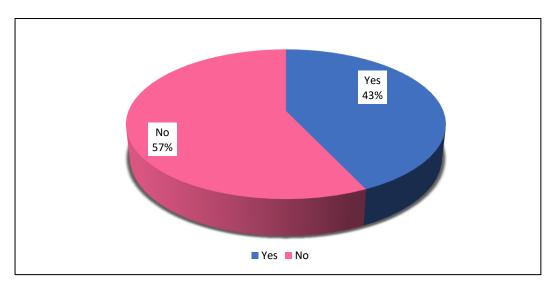


Figure 7. Scenario of the raw eggs close to other products such as vegetable

## 4.2 Assessment of hygiene practices of table eggs

## 4.2.2. Collection, types and grading system of table eggs

The Survey discovered that eggs are collected from different sources. 52% (figure 8) Vendors collect eggs from supplier whereas 30% from dealer and 18% from others. Among the collected eggs, chicken (layer) eggs are 46%, chicken (deshi) eggs only 6% (figure 9) multi species (Chicken, Duck and Quail) eggs are 48%.

The conclusion drawn from this survey is that rearing a variety of eggs (both dirty and clean) may be a factor in the spread of the disease.

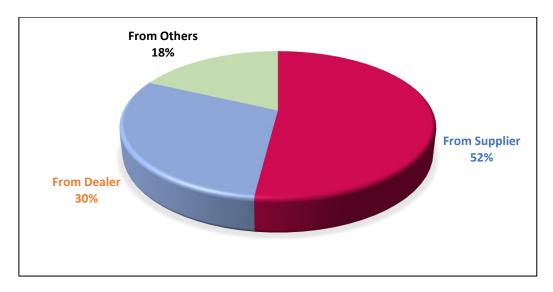


Figure 8. Collection of eggs

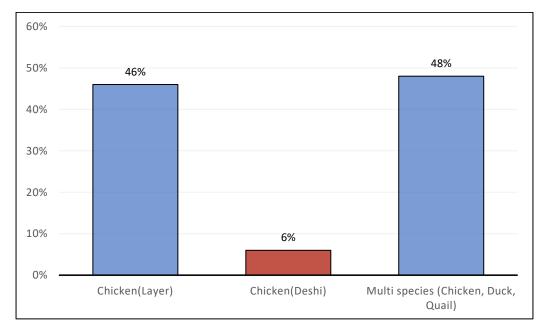


Figure 9. Types of eggs

## Grading of eggs

The purpose of the egg grading is to sort the eggs into classes based on exterior factors which includes cleanliness and soundness of the shell and the interior factors such as albumen, yolk, air mobile and possible abnormalities. The present survey findings also found that only 36% (figure 10) vendors maintain the grading system of egg where most of the vendors didn't grade the eggs. So, it can be said that the egg market of Bangladesh, a developing country are still not commercialized properly and invisibly it's hampering the marketing system.

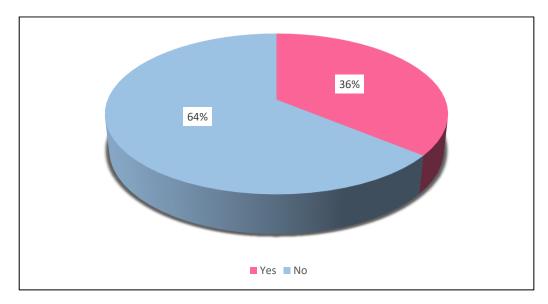


Figure 10. Grading of eggs

The present findings are similar to another research of Tamiru *et al.* (2019), revealed that in developed countries eggs are graded before providing to market; However, such practices are not followed in developing countries like Ethiopia.

# 4.2.2 Transportation, storage and Packaging system of table eggs

From Survey findings, figure11 notified that 74% vendors used van, 16% vendors used both van and rickshaw, 6% vendors used truck and 4% used others for egg transportation. According to USDA, the US department of commerce's sanitary food transportation, Act of 1990 requires vehicles to be used exclusively for transporting food. The ambient temperature of shell eggs packed for consumption, storage and shipping under refrigerated conditions does not exceed 45°F (7.2°C). From this survey, it was found that in the absence of mandatory safety rules for the transport of eggs and there is no reference temperature inside the van or truck, the contamination can transfer from one egg to another.

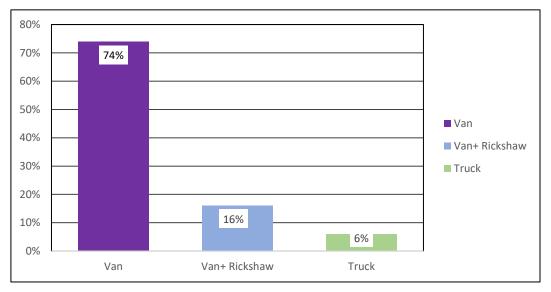


Figure 11. Transportation of eggs

# Storage condition and time

The result also revealed that 32% (figure 12) vendors stored the egg inside the shop with ceiling fan where most of the vendors (68%) stored the egg inside the shop without ceiling fan. Vendors usually sold their eggs within 3 days whereas some vendors kept the eggs more than 3 days (26%). So, it can be said there have a great demand of table eggs to the consumers. From this survey it also revealed that egg stored in shop showed better quality might be the vendors do not kept eggs more than 3 days.

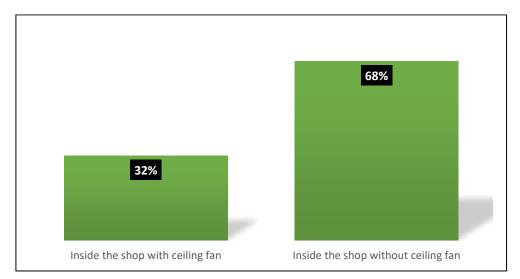


Figure 12. Storage condition

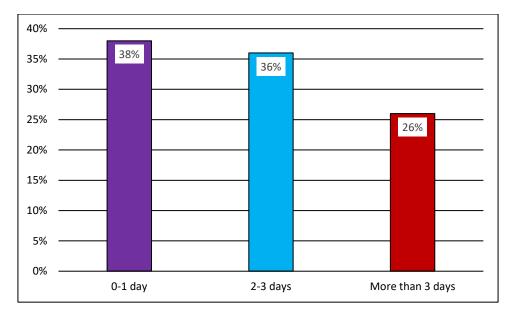


Figure 13. Storage time

The result of the findings are similar with the previous study of Akter *et al.* (2014), found that there was no significant effect of storage temperature on shell thickness, when the eggs were held at room and the shell thickness of eggs held at two different temperatures (4 and 28-31 °C) were not affected by the storage temperature. Yolk color was unchanged during the whole storage period; however, eggs stored in refrigerator showed better quality up to 28 days and in room temperature up to 14 days.

Findings of the present study are in agreement with Çağlayan *et al.* (2009) and Dudusola (2009), who reported that the storage time and temperature had no effect on shell thickness of partridges and Japanese quail eggs, respectively.

## Packaging of eggs

The survey also showed that (figure 14) maximum vendors used both polythene and case (36%) in their shop rather used it singly and they also used paper packet for packaging of eggs.

Eggs are best packed in small cardboard containers and covered with plastic. This type of packaging system has been already appeared in some company such as Kazi, CP etc. But small quantities were found in the wet market. The survey revealed that this low packaging system (polythene, paper or dirty cases) might be cause of the egg contamination.

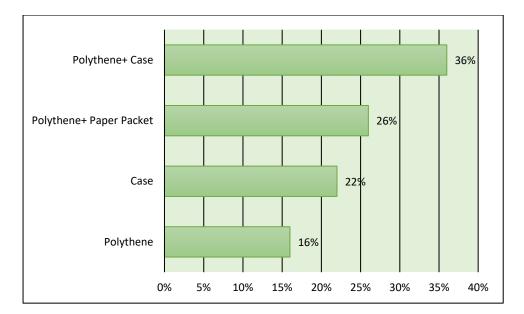


Figure 14. Packaging of eggs

## 4.3 Keeping the record and disposal process of broken, dirty and rotten egg

The Survey disclosed that (figure 15) only 32% vendors kept the record of broken, dirty and rotten eggs where 68% vendors never kept the record. It also revealed that (figure16) only 6% vendors dumped the broken eggs into drain and garbage whereas most of the vendors (94%) sold the broken eggs to customer, hotel and restaurant. In the context of above result it can be said that most of the vendors are not conscious about the food safety.

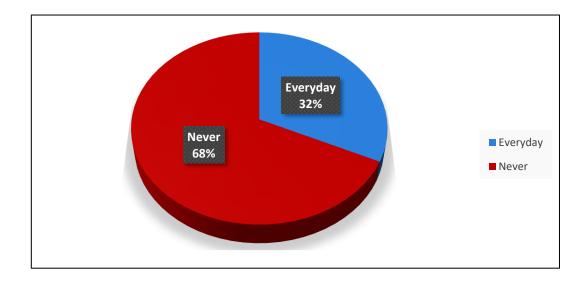


Figure 15. Recording keeping

It also notified from the survey that most of the vendors sold the broken eggs to hotel, customer and restaurants at a lower price.

The present result are in similar with the previous research of Edema and Atayese, (2006), found that in developing countries like Nigeria, they sorted and sold at about half the prices of whole un cracked eggs. These cracked eggs should not be marketed in any establishment, since they do not comply with the legislation, as they pose a risk to public health. Lima *et al.* (2018), pointed that that cracked eggs are suitable for penetration of microorganisms, which can lead to the contamination of those who consume them.

The present findings showed that cracked eggs have higher demand as they are available at the price Tk 2-3 piece where the regular eggs are sold at Tk 6-10 each. These activities of the egg vendor's support to more chance of outbreak of foodborne disease.

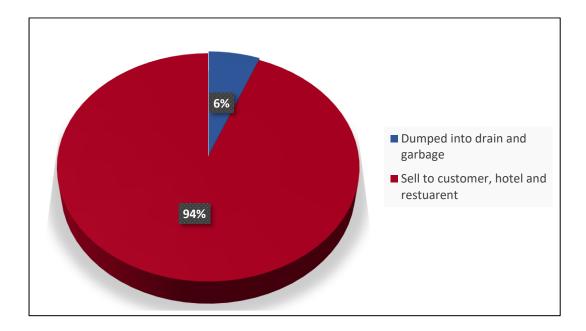


Figure 16. Disposal of broken eggs

This survey exposed that in case of rotten eggs, maximum vendors (58%) dumped the eggs into the garbage, where 20% vendors dumped into the drain, 14% vendors used both drain and garbage and only 8% vendors back the eggs into warehouse (figure 17). So, from the above findings, it can be concluded that vendors were not well concern about record keeping and most of the vendors further sold the broken eggs rather dumping them.

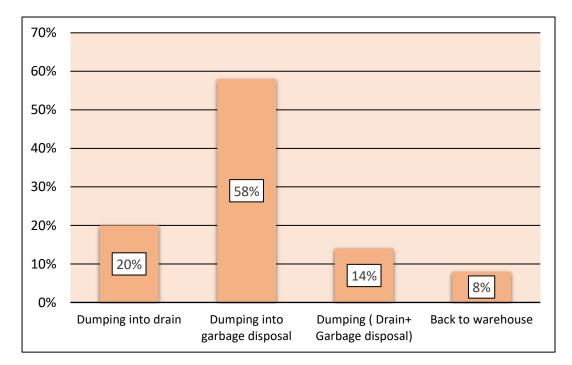


Figure 17. Disposal of rotten egg

# **Microbiological Analysis**

## 4.4 Total salmonella count

The values of the total salmonella count of egg samples are recorded in Table 4.1. The average counts of bacteria in sample area of clean eggs and dirty eggs are given below .The minimum and maximum ranges of salmonella count as revealed in chicken eggshell surface in dirty eggs were log 6.04 ( $1.1 \times 10^6$  CFU/ml) to log 6.3 ( $2.00 \times 10^6$  CFU/ml); in clean eggs, log 4.75 ( $5.66 \times 10^4$  CFU/ml) to log 5.00 ( $1.02 \times 10^5$  CFU/ml), (Table 2). The highest bacterial load for dirty eggs was found in Taltola Market and lowest was found for dirty eggs in SAU Market. For clean eggs, the lowest bacterial load was found in SAU Market & highest was found in Krishi Market.

Market name	Dirty eggs	Clean eggs	Standard value	
-	Average counts of bacteria(log 10	Average counts of bacteria(log 10		
	value)	value		
SAU Market	$1.10 \times 10^{6}$	$5.66 \times 10^{4}$		
	(6.04)	(4.75)		
Agargoan Market	$1.30 \times 10^{6}$	$9.5 \times 10^{4}$		
0 0	(6.11)	(4.97)		
Taltola Market	$2.00 \times 10^{6}$	$7.0 \times 10^{4}$	Not detected in 25g	
	(6.3)	(4.84)		
Krishi Market	$1.65 \times 10^{6}$	$1.02 \times 10^{5}$		
	(6.23)	(5.00)		
Bihari Kamp market	1.20×10 <sup>6</sup> (6.07)	6.0×10 <sup>4</sup> (4.77)		

## Table 2. Counts of TSC in different egg sample

Source: FSAI (2001).

## \*All counts are expressed in Colony Forming Units (CFU)

#### 4.5 Total coliform count

The values of the total coliform count of egg samples are recorded in Table 4.2. The average counts of bacteria in sample area of clean eggs and dirty eggs are given below .The minimum and maximum ranges of coliform count as revealed in chicken eggshell surface in dirty eggs were log 6.0 ( $1.0 \times 10^6$  CFU/ml) to log 6.34 ( $2.20 \times 10^6$  CFU/ml); in clean eggs, log 4.6 ( $4.0 \times 10^4$  CFU/ml) to log 5.11 ( $1.30 \times 10^5$  CFU/ml), (Table 3). The highest bacterial load for dirty eggs was found in Krishi Market and lowest was found for dirty eggs in Agargoan Market. For clean eggs, the lowest bacterial load was found in SAU Market & highest was found in Bihari Camp Market.

Market name	Dirty eggs Clean eggs		Standard value	
	Average counts of bacteria(log 10 value)	Average counts of bacteria(log 10 value	-	
SAU Market	1.60 ×10 <sup>6</sup> (6.20)	4.0×10 <sup>4</sup> (4.6)		
Agargoan Market	1.00 ×10 <sup>6</sup> (6.00)	9.0×10 <sup>4</sup> (4.95)	10 or less per organism	
Taltola Market	1.08×10 <sup>6</sup> (6.03)	$1.2 \times 10^{6}$ (5.07)		
Krishi Market	2.20×10 <sup>6</sup> (6.34)	8.0 ×10 <sup>4</sup> (4.9)		
Bihari Camp market	$1.80 \times 10^{6}$ (6.25)	1.30×10 <sup>5</sup> (5.11)		

# Table 3. Counts of TCC in different egg sample

Source: QFCIAF, 1995

\*All counts are expressed in Colony Forming Units (CFU)

# 4.6 Prevalence of *Salmonella* spp. and *E.coli* in different wet market of Sher-e-Bangla Nagar area

Overall of 50 egg samples were gathered from different wet of Sher-e-Bangla Nagar area of Dhaka city. A total of 50 sample tested, 15 (30%) samples tested positive for *Salmonella* and 21 (42%) samples tested positive for *E.coli*. (Table 4.3) the presence of the bacteria is higher in dirty eggs compared to the clean eggs. The lowest contamination rate is 60%, which is found in SAU Market & Taltola Market and highest bacterial contamination rate is 90%, which was found in Krishi Market.

Sl No. Market name		No. of Collected	No. of samples positive for <i>Salmonella</i> spp.		No. of samples positive for <i>E. coli</i>		Prevalence %
		Samples	Dirty eggs	Clean eggs	Dirty eggs	Clean eggs	-
1	SAU Market	10	1	1	2	2	60%
2	Agargoan Market	10	2	1	3	1	70%
3	Taltola Market	10	2	1	2	1	60%
4	Krishi Market	10	2	1	4	2	90%
5	Bihari Kamp market	10	3	1	3	1	80%
	Total	50	1	.5		21	

## Table 4.3 Prevalence of Salmonella and E.coli in different egg sample

The aim of this present study was to detect the presence of bacteria (*Salmonella and E. coli*) on the eggshell surface. *Salmonella* spp. are capable of growing in foods at room temperature has been reported by Phillips *et al.* (1947) and Hall *et al.* (1967), but probably would be unable to grow in properly refrigerated foods. Present study has demonstrated an overall prevalence (30%) of *salmonella* spp. contamination where the lowest was in eggshell sample of clean egg and the value was log  $4.75(5.66 \times 10^4 \text{ CFU/ml})$  and highest was in dirty eggshell sample and the value was log  $6.3 (2.00 \times 10^6 \text{ CFU/ml})$ . The highest bacterial load for dirty eggs was found in Taltola Market and the lowest bacterial load was found in SAU Market.

In this study, the prevalence of *Salmonella* spp. for table eggs from the selective market it was 30%, with the highest prevalence being 40.00% in the Bihari Camp market and the lowest prevalence being 20% in the SAU market. The result of this study was approximately similar with the findings of Rezaul *et al.* (2017), a survey conducted in Savar of Dhaka that 28% of eggshell were positive for *salmonella* spp. The present study was contradictory with other study conducted in developed countries like China, France, Canada, Australia etc. Li *et al.*, (2020) found in their study that only 4.4 % prevalence of *Salmonella* spp. in wet market of China. Recent study in France showed that prevalence of salmonella in the egg shells was 0.3 -1.05 % (Chemaly *et al.*, 2009). Other study in Belgium, New Zealand, Australia, and Canada reported a range of 2-13% *Salmonella* infection in large sampling frame as part of public health surveillance system .(Murchie *et al.*, 2008 and Gould *et al.*, 2004)

This analysis found few colonies that were below the acceptable range, or sometimes no colonies were found in several eggshell samples that were within the standard level. The result of this analysis shows that the eggs have been mistreated at every stage and the eggs should be stored at room temperature (23-30°C) for a several days before sale. Then the bacteria would not only need a short time to get to a level to multiply, at which point they could produce significant amounts of enterotoxin. The presence of multiple Salmonella microbes in eggs has the potential to cause foodborne illness in eggs. This type of bioburden indicates contamination of eggs with Salmonella spp. and contamination can occur at any stage of production such as collection, transport, and marketing. This higher prevalence and bacterial load were probably due to poor sanitary conditions in the wet market. Therefore, the reuse of dirty trays is a potential source of *Salmonella* spp. contamination in developing countries like Bangladesh.

In this analysis, total 21 samples of two categories (clean eggs and dirty eggs) exceeded the coliform unit of the standard level. The lowest was found in eggshell samples of clean egg and the value was log  $4.6(4.0 \times 10^4 \text{ CFU/ml})$  and the highest was found in eggshell samples of dirty egg and the value was log 6.34 ( $2.20 \times 10^6 \text{ CFU/ml}$ ). Out of 50 samples, 21 samples (42%) were found to be positive for *E.coli*. The results of TCC for 29 samples (clean eggs and dirty eggs) were nil (absence of coliform) also indicate standard level. So these samples also considered as safe egg.

Many research workers while examining foods of various types believe that a minimum level of coliform contamination will always be present (Lewis and Angelotti, 1964). The exact significance of the association of these organisms although is more or less understood, but the sanitarians consider coliform counts as an indicator of fecal pollution (Hall et al. 1967). Recommended limits for food samples, raw meat sand some milk products are in the range of 10 or < 10 coliform organism per gm (Quartermaster Food and Container Institute of the Armed Forces, MIL-M-13 966, 1955). The coliform index as an index of sanitary quality is applicable to at least some foods. It can be seen that low numbers of coliform are permitted ranging from 1 to not over 100/g or ml. Implicit in these standards are answers to questions of feasibility and safely (Slanetz *et a*1., 1962).

This study showed that the presence of coliform bacteria in the contaminated egg samples was too high and did not meet the microbial standards or limits. Eggs can be contaminated by dust, feces and other dirty products during handling or transport. The available data showed that the highest incidence of coliform bacteria in the eggs indicated unhygienic conditions, which revealed the fact that the eggs were contaminated with facial material, dust, etc. In this study, the prevalence of *E.coli* for table eggs from the selective market it was 42%, with the highest prevalence being 60.00% in the Krishi market and the lowest prevalence being 30% in the Taltola market. The result of this study was nearly similar with the findings of Hossain *et al.* (2021), a survey conducted in Rajshahi that 41.66% of eggshell were positive for *E.coli*. The results of this experiment are more or less related to the studies presented previously. The higher prevalence of coliform bacteria in eggs underlines the public health importance.

## **CHAPTER 5**

## **CONCLUSION AND RECOMMENDATION**

This chapter summarizes key findings and draws conclusions. The study mainly focuses on the evaluation of vendor hygiene practices in various wet markets of Sher-e-Bangla Nagar, North City Corporation. The specific purpose of this analysis was to investigate the hygienic practices of vendors in wet markets during egg handling, storage, transportation, distribution, etc. and to identify the pathogen (*Salmonella* spp. *and Escherichia coli*) on the eggshell surface. The research process was divided into two parts; surveys with a questionnaire and microbiological analysis of the eggshell surface.

Findings from the experiment showed that eggs sold are highly contaminated due to poor handling by the vendors, storage and environmental conditions. The survey notified that only 4% vendors wash their hands before and after handling of eggs where 96% vendors didn't wash hands before and after handling of eggs.

It also revealed that 37% vendors clean their eggs before selling and 63% vendors do not clean their eggs before selling. So, there had a great chance of spread the diseases if the eggs are contaminated. Even only 36% vendors maintain the grading system of egg where most of the vendors didn't grade the eggs.

The results showed that 32% vendors kept the record of broken, dirty and rotten eggs where 68% vendors never kept the record. Having another important survey showed that most of the vendors (94%) sold the broken eggs to customer, hotel and restaurant.

It also found from the survey that *Salmonella* spp. and *E. coli* was present 30% and 42% in egg samples.

From microbiological analysis, it was revealed that the highest bacterial load for dirty eggs was found in Krishi Market and lowest was found for dirty eggs in Agargoan Market in coliform counting. For clean eggs, the lowest bacterial load was found in SAU Market & highest was found in Bihari Camp Market.

In salmonella counting method, it also found that the highest bacterial load for dirty eggs was found in Taltola Market and lowest was found for dirty eggs in SAU Market. For clean eggs, the lowest bacterial load was found in SAU Market & highest was found in Krishi Market.

In the context above the results, it can be said that the bacterial load was found higher in dirty eggs rather than clean eggs. Though eggs were highly contaminated with bacteria like *Salmonella* spp. and *E.coli*, which is major concern in poultry industry as well as public health.

Based on this study, it was concluded that edible eggs from different wet markets of Sher-e-Bangla Nagar area of North City Corporation were contaminated with *Salmonella* spp. and *Escherichia coli* organisms.

The study indicated that the increase in outbreaks of the organism may be due to unsanitary conditions, reuse of dirty trays and improper handling of eggs during egg collection, marketing and storage.

Contamination of eggs due to mistreatment by sellers and customers can cause morbidity and mortality in the population, especially in developing countries such as Bangladesh. It also notified that egg sellers' knowledge of hygiene practices was disappointing. Most vendors had lower knowledge of hygiene practices, while some still have several safe hygiene practices.

On the basis of findings and conclusion, it may be recommended that attempts should be taken to make awareness among the vendors by regular training and provide food safety guide for increasing knowledge on good hygienic practice. It also recommended that wet markets must be regularly visited by food inspectors to monitor the hygienic environment.

It should be remembered that equipment's that are mainly used for handling and storing of eggs should be always cleaned. Vendors should maintain proper keeping quality of eggs prior to sale. They should store and sell their eggs under refrigerator or good condition to reduce microbial load.

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## **APPENDICES**

# "Hygiene Practices of Table Eggs at Different Wet Market in Sher- e-Bangla Nagar Area of Dhaka North City Corporation: A Public Health Concern"

Dept. of Animal Nutrition, Genetics & Breeding, SAU

## Appendix -I

#### Questionnaire for field survey

#### **\*** Basic information about vendors:

- 1. Name:
- 2. Education level:
- 3. Store name:
- 4. Market name:
- 5. Area:
- 6. Mobile number:

## **\*** Vendors concern about food hygiene:

- 1. Do you know about the consumption of raw and undercooked egg may be harmful to health?
  - □ Yes
  - □ No
- 2. Do you wash your hands before and after handling of eggs?
  - $\Box$  Yes
  - □ No
- 3. Do you know about the dirty, broken egg related disease?
  - $\Box$  Yes
  - □ No
- 4. Do you keep the raw eggs close to other products such as vegetable?
  - □ Yes
  - □ No
- 5. Do you clean eggs before selling?
  - $\Box$  Yes
  - 🗆 No
- 6. If you clean your eggs before selling, what is the cleaning procedure?
  - $\Box$  Dry wash
  - $\Box$  Wet wash

## ✤ Information about the selling pattern of table eggs:

- 1. Collection of eggs:
  - a. Any company (specify .....)
  - b. Dealer
  - c. Supplier
  - d. Others (.....)
- 2. Which type of eggs are collected:
  - a) Chicken indigenous/farm egg
  - b) Duck
  - c) Quail
- 3. Do you follow any grading system of eggs?
  - □ Yes
  - 🗆 No
- 4. Grading system:
  - a) A grade
  - b) B grade
  - c) Dirty
  - d) Cracked
  - e) Size
  - f) Others (.....)
- 5. Transportation process:
  - a) Truck
  - b) Van
  - c) Rickshaw
  - d) Others (....)
- 6. Storage of eggs:
  - a) Inside the shop with ceiling fan
  - b) Inside the shop without ceiling fan
  - c) Others (....)
- 7. Time duration:
- 8. Packaging system:
  - a) Paper packet
  - b) Polythene bag
  - c) Nylon bag
  - d) Cage
  - e) Others

- 9. Any expiration date:
  - a) Yes
  - b) No

# Information about the disposal process of dirty, broken and rotten eggs:

- 1. Keeping record of broken, dirty and rotten eggs:
  - a) Daily
  - b) Weekly
  - c) Monthly
  - d) Never
- 2. Disposal procedure of rotten eggs:
  - a) Dumping into drain
  - b) Dumping into garbage disposal
  - c) Use as compost
  - d) Others (....)
- 3. Disposal process of broken egg:
  - a) Sell into the hotel or restaurants
  - b) Sell to the customers
  - c) Dumping into the drain
  - d) Others (....)

# Appendix-II

# (List of sampled vendors)

SL.NO	VENDORS NAME	STORE NAME	MARKET NAME	MARKET AREA	MOBILE NO.
1	Abdur Rahman	Perfect Confectionary	Paka Market	Agargoan	01812-087491
2	Mizanur Rahman	Mizan Store	Paka Market	Agargoan	01012 007 171
3	Abdul		Agargoan	Agargoan	01675406113
4	Akbar Hossein		Paka Market	Agargoan	01670551951
5	Hasib		Agargoan	Agargoan	01822-500850
6	Billal		Agargoan	Agargoan	01022 000000
е 7	Azhar Ali		Taltola	Agargoan	
8	Anwar		Taltola	Agargoan	
9	Billal	Billal store	Batzor	Sher-e-Bangla Nagar	01973189436
10	Munna	Munna Store	Mohammadpur	Mohammadpur	01714643247
11	Shamol	Shadesh Store	Mohammadpur	Mohammadpur	01727111001
12	Jahangir	Jahangir Store	Agargoan	Sher-e-Bangla Nagar	01849515180
13	Raihan	Raihan store	Agargoan	Sher-e-Bangla Nagar	01772058301
14	Sajal	Sajal Store	Agargoan	Sher-e-Bangla Nagar	01730748553
15	Abdur Rajjak	Rajjak Store	Agargoan	Sher-e-Bangla Nagar	01716142868
16	Abdullah		Agargoan	Sher-e-Bangla Nagar	01716142868
17	Nasir	Nasir store	Agargoan	Sher-e-Bangla Nagar	01893929898
18	Saiful		Agargoan	Sher-e-Bangla Nagar	01959287755
19	Titu		Agargoan	Sher-e-Bangla Nagar	01876666986

# Appendix-II (CONT'D)

SL.NO	VENDORS NAME	STORE NAME	MARKET NAME	MARKET AREA	MOBILE NO.
20	Anik		Agargoan	Sher-e-Bangla Nagar	01521-369841
21	Abdul Malek	New Bismillah Broiler and Egg Store	Krishi Market	Mohammadpur	01886490464
22	Md.Arif Hosen	B-baria Egg store	Krishi Market	Mohammadpur	01837-904025
23	Parvez	New Kacha Bazar Broiler House		Mohammadpur	01402751223
24	Anwar	Bismillah Depertmenatal Store	Krishi Market	Mohammadpur	01308820984
25	Md. Jakir	Chadpur Broiler House	Krishi Market	Mohammadpur	01792-976696
26	Md. Humayan	Janata Broiler	Krishi Market	Mohammadpur	01837-560819
27	Rjib Alam	Kacha Bazar Broiler House	Krishi Market	Mohammadpur	01717489280
28	Abdul Mannan		Mini Market	Sher-e-Bangla Nagar	01756826846
29	Lutfur Rahman	Mayer Doa	Camp Bazar	Mohammadpur	01921-491433
30	Junayed	Junayed Store	Camp Bazar	Mohammadpur	
31	Jubayer	Ma Store	Camp Bazar	Mohammadpur	
32	Gias uddin	Sakal Sondha Egg Store	Camp Bazar	Mohammadpur	
33	Hasan	Hasan Store	Camp Bazar	Mohammadpur	
34	Iyub	Iyub Store	Camp Bazar	Mohammadpur	
35	Rakib		Camp bazar	Mohammadpur	
36	Abu Bokkor Siddik	Barishal chicken house	Taltola	Agargoan	01674604376
37	Ibrahim Khalil	Noakhali treaders	Taltola	Agargoan	01731788226
38	Jewel	Bismillah Variety Store	Taltola	Agargoan	

# Appendix-II (CONT'D)

SL.NO.	VENDORS NAME	STORE NAME	MARKET NAME	MARKET AREA	MOBILE NO.
39	Adul Karim	Keraniganj General store	Taltola	Agargoan	
40	Ismail vuiya	Vuiya General store	Taltola	Agargoan	
41	Abu Bakar Siddik		Taltola	Agargoan	
42	Reza	Padma Broiler House	Krishi Market	Mohammadpur	01962-064149
43	Mujibur Rahman	Modija General Store	Krishi Market	Mohammadpur	01712-539565
44	Masum		Krishi Market	Mohammadpur	01722-92508
45	Rakib		BNP Market	Sher-e-Bangla Nagar	01778-684923
46	Abdur Rahman	Abdur rahman tea store	BNP Market	Sher-e-Bangla Nagar	01986210088
47	Shafiqur Rahman		BNP Market	Sher-e-Bangla Nagar	01711281313
48	Badal	Mayer Doa	Mini Market	Sher-e-Bangla Nagar	01883265475
49	Sajib	Mim General Store	Krishi Market	Mohammadpur	01811-270549
50	Hannan	New Janata Broiler	Krishi Market	Mohammadpur	01747-227856