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Comparative Test on the Level Contamination of *Escherichia coli* and *Salmonella sp.* on Broiler Meat at Slaughterhouses and Traditional Markets in Medan

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Abstract. Contamination of meat is common Contamination of meat often occurs which endangers the health of consumers. This study aims to determine the presence of bacterial contamination of *Escherichia coli* and *Salmonella sp.* on broiler meat at slaughterhouses and traditional markets in Medan City. This study used a survey method with a purposive sampling technique and used 60 samples (30 from slaughterhouses and 30 from traditional markets). There were 3 slaughterhouses (slaughterhouses Johor, slaughterhouses setia budi, and slaughterhouses padang bulan) and three traditional markets (traditional market Johor, Setia Budi and Padang bulan). Samples were analyzed at the Laboratory Microbiology Faculty of Mathematic and Natural Science Universitas Sumatra Utara Medan. The results showed that the average total amount of *Escherichia coli* in the sample from chicken slaughterhouses was 3.15 log CFU/g, and from the traditional markets was 3.39 log CFU/g. The number of samples contaminated by *Salmonella sp.* in chicken slaughterhouses was 17 of 30 samples, whereas on the traditional market were 20 of the 30 samples. This study concludes that the average total amount of *Escherichia coli* microbes in samples from chicken slaughterhouses and traditional markets in Medan City all of them had exceeded the maximum limit of microbial contamination, based on the microbiological quality requirements of SNI 3932: 2008, which is 1×10^1 CFU/g. While the bacterial contamination rate of *Salmonella sp.* in chicken slaughterhouses was 56%, and in the traditional market was 66%.

Keyword: Bacterial, Chicken slaughterhouses, Microbiological quality,SNI, Traditional market

1 Introduction

Chicken meat is a popular food source for the community. This fact is accompanied by the large amount of chicken meat consumed by the people of Indonesia, which is 0.538 kilos/per capita/month and continues to increase based on a survey by the Central Statistics Agency (BPS). Chicken meat is a livestock product with properties and characteristics that are very sensitive to

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damage due to contamination [1]. Chicken meat is a food ingredient that has perishable food properties, so it requires attention so that its quality is always maintained. Chicken meat has a high protein content, so it is susceptible to damage [2]. High protein content and postharvest handling also make chicken meat susceptible to microbial growth [3]

The chemical composition of chicken meat consists of 74.86% water content, 23.20% protein, 1.65% fat, 0.98% minerals, and 114 kcal calories. The pH value of chicken meat after 24 hours (post-mortem) is 5.5- 5.9. Chicken meat is a source of animal protein rich in calories, fat, vitamins and minerals [4] Initial problems with chicken meat often occur due to biological factors, namely the discovery of Escherichia coli and salmonella sp., which is beyond the threshold of the Indonesian National Standard (SNI: 3932: 2008). An increase in the number of bacteria beyond the threshold can cause chicken meat to experience damage to its nutritional value and a decrease in quality due to the process of decay by bacteria. Hence, the meat is unfit for consumption [5]. Microorganism contamination conditions outside the threshold can potentially cause dangerous diseases if consumed by humans, often called foodborne diseases. Therefore, Escherichia coli bacteria in chicken meat must be known to ensure the feasibility of chicken meat for consumption [6].

In the process of slaughtering chickens, both at the Chicken Slaughterhouse (TPA) and Traditional Markets, they are prone to contamination by Escherichia coli and Salmonella sp. Factors that can trigger contamination usually occur when handling chicken slaughter, which is not good; the cutting location is dirty, and the equipment used in the cutting process can cause contamination. Research on Escherichia coli detection conducted by [7] on chicken meat samples taken along the distribution chain in Gowa Regency showed (100%) of 24 meat samples contaminated with Escherichia coli bacteria and exceeded (BMCM) and Salmonella sp. chicken meat conducted by [8] who took samples from two different locations obtained a percentage of 55%, namely from traditional markets as many as 17 out of 40 samples (42.5%) while from supermarkets obtained 5 out of 40 samples (12.5%).

Based on the explanation above, it is necessary to research to detect the level of contamination by *Escherichia coli* and *Salmonella sp.* on various cuts of broiler chickens in Medan. Information about the presence of *Escherichia coli* and *Salmonella sp.* on chicken meat products in Medan City will increase the Medan people's awareness in buying and consuming chicken meat in Medan City.

2   Materials and Methods

Materials

The research was carried out at chicken slaughterhouses (Johor, Setia Budi and Padang Bulan) and traditional markets (Johor, Setia Budi and Padang Bulan), followed by examinations at the

USU Faculty of Mathematics and Natural Sciences Microbiology Laboratory. This research was conducted in May–June 2022.

The materials used in this study were 60 samples of chicken meat, with 25 grams of each sample taken from traditional chicken slaughterhouses and markets. The media for testing were a solution of Buffered Peptone Water (BPW) Sigma-Aldrich, Eosin Methylene Blue Agar (EMB) Sigma-Aldrich, Salmonella Shigella Agar (SSA) Merck, Aquadest, 70% alcohol, cotton, and tissue.

The tools used in this study were stationery, sterile plastic for packing samples, a freezer, a spatula, test tubes, Erlenmeyer flasks, test tube racks, Bunsen burners, Petri dishes, incubators, autoclaves, sterile cabinets, ice boxes, analytical scales, boxes cooler, marker, gloves, mask and lamp, pestle and mortar, tip and micropipette, bunsen, matches, marker, cling wrap, aluminium foil, hot plate, vortex, measuring cup, magnetic stiver.

Methods

This research is a survey study of meat chicken from slaughterhouses and traditional markets to determine the presence of *Escherichia coli* and *Salmonella sp*. Information about the condition of broiler chickens was obtained by direct observation at the time of sampling. The data collected were related with the cleanliness of the place of sale, the cleanliness of the equipment used, the condition of the seller/handler in general and the environment around the place of sale. Samples analyzed at the Laboratory Microbiology Faculty of Mathematic and Natural Science Universitas Sumatera Utara Medan.**Data analysis**

Data is presented as a frequency distribution table to see the presence of *Salmonella sp*. contained in broiler chicken meat and continued with the Chi-Square test (χ^2) with statistical software. The Chi-Square test is faced as a test of whether the difference between the observed frequency and the expected frequency of a limited sample is significant or not [9]

Meanwhile, to see the difference in the number of *Escherichia coli* found in fresh chicken meat at Chicken Slaughterhouses (Johor, Setia Budi and Padang Bulan) and Traditional Markets (Johor, Setia Budi and Padang Bulan), is continued with a different t-test (partial test /individual) with statistical software. According to [10], a different test t-test is used to test how far the independent variables used in this study are affected individually by partially explaining the dependent variable.

Sample Examination for *Escherichia coli* and *Salmonella sp.* (SNI:2897:2008)

The initial stage was carried out, namely the preparation of the meat sample before analysis by grinding the samples taken and weighing first as much as 25 grams and then adding 225 ml of BPW solution in the stomacher for 2 minutes. Then pour 9 ml of the BPW solution into a sterile test tube and add 1 ml of the 10^{-1} dilution suspension with a sterile pipette into the 9 ml BPW solution to get a 10^{-2} dilution. Make a 10^{-3} and 10^{-4} dilution similarly. Then 1 ml of the suspension

from each dilution was put into a sterile petri dish. Then the EMBA and SSA media were cultured in a petri dish for 24 hours.

a. Making Buffered Peptone Water (BPW) Medium

A total of 4.5 grams of Buffered Peptone Water medium was weighed, put into a 250 mL Erlenmeyer then, added 225 mL of distilled water was and stirred until homogeneous. The medium was sterilized using an autoclave at 121^0C with a pressure of 15 psi/2 atm for 15 minutes.

b. Making Eosin Methylene Blue Agar (EMB) Medium

A total of 16.2 grams of Eosin Methylene Blue Agar medium was weighed, put into a 500 mL Erlenmeyer then, added 450 mL of distilled water was and stirred until homogeneous. Heated on a hotplate until boiling. The medium was sterilized using an autoclave at 121^0C with a pressure of 15 psi/2 atm for 15 minutes.

c. Preparation of Salmonella Shigella Agar (SSA)

A total of 27 grams of Salmonella Shigella Agar medium was weighed, put into a 500 mL Erlenmeyer then, added 450 mL of distilled water was and stirred until homogeneous. Heated on a hot plate until boiling. The medium was sterilized using an autoclave at 121^0C with a pressure of 15 psi/2 atm for 15 minutes.

d. Dilution

After the BPW solution is prepared, weigh 25 grams of chicken meat sample, add 225 ml of sterile 0.1% BPW solution into a sterile bag containing meat, and then homogenize with a stomacher for 1 to 2 minutes. Then carry out the dilution process from sample destruction to a 10-1 dilution to a 10-4 dilution. Multilevel dilution aims to minimize or reduce the number of microbes suspended in the liquid. A ratio of 1: 9 is used for the sample and the first and subsequent dilutions so that the next dilution contains 1/10 of the microorganism cells from the previous dilution.

e. Escherichia coli Contamination Testing

A total of 25 grams of chicken meat was weighed and put into 225 mL of BPW solution. Then serial dilutions were carried out 10^{-1} - 10^{-4}. Each 1 ml of each dilution was cultured using the pour plate method on Eosin Methylene Blue Agar medium. Incubated for 24 hours. Observed and counted. Metallic green E.coli colonies on Eosin Methylene Blue Agar medium.

f. Salmonella Contamination Testing

A total of 25 grams of chicken meat was weighed and put into 225 mL of BPW solution. Then serial dilutions were carried out 10^{-1} - 10^{-4}. Every 1 ml of each dilution was cultured on Salmonella Shigella Agar medium. Incubated for 24 hours. Observe and count the amount. Salmonella colonies are colourless, transparent and have a black precipitate.

3 Result and Discussion

Bacterial Analysis of *Escherichia coli*

The results of the analysis of *Escherichia coli* in broiler meat samples marketed at chicken slaughterhouses and traditional markets in Medan City can be seen in Table 6 below.

Table 6. Number of *Escherichia coli* bacteria (Log CFU/g) at chicken slaughterhouses and traditional markets in the city of Medan

Code	Number	Value Log CFU/g	
		Slaughters	Traditional Market
1	1	3.00	4.00
	2	2.17	3.00
	3	4.30	3.30
	4	3.00	2.17
	5	2.17	3.00
	6	3.30	4.30
	7	4.00	3.00
	8	3.30	4.30
	9	3.00	3.39
	10	2.17	2.17
2	1	3.00	4.30
	2	2.17	4.00
	3	3.30	2.69
	4	3.17	4.00
	5	4.30	2.17
	6	3.17	4.30
	7	4.00	4.47
	8	2.17	3.00
	9	3.30	4.47
	10	4.00	3.17
3	1	3.17	3.00
	2	3.00	4.30
	3	2.17	3.30
	4	3.00	3.69
	5	3.30	4.00
	6	3.17	3.39
	7	3.00	2.17
	8	3.17	2.39
	9	4.30	4.00
	10	3.30	2.39
Average*		3.15	3.39

Note :*= t value < t table is not significantly different (P> 0.05)

Table 6 shows that the average amount of *Escherichia coli* contamination in chicken slaughterhouses is 3.15 log CFU/g and in traditional markets with an average of 3.39 log CFU/g. Contamination at traditional markets can be caused by unhygienic handling, stacked meat, storage conditions without refrigeration and being in the open air [6]. Based on SNI 3932: 2008, the maximum limit for microbial contamination (BMCM) of Escherichia coli on the microbiological quality requirements for meat is 1×10^1 Colony Forming Units per gram (CFU/g) [7]. The results of the analysis from Table 6 showed that of the 30 samples of broiler chicken meat from chicken

slaughterhouses and 30 samples of broiler chicken meat from traditional markets analyzed showed Escherichia coli testing in research conducted in the laboratory; there were 60 chicken meat samples (100%) that exceeded the maximum limit of microbial contamination of chicken meat bacteria contaminated with Escherichia Coli bacteria.

These results are the same as research conducted by [7] on chicken meat samples taken along the distribution chain in Gowa Regency, showing (100%) of 24 meat samples were contaminated with Escherichia coli bacteria and exceeding (BMCM). Research by [11] showed that meat samples in Gorontalo City consisting of 13 samples and nine samples (69%) had exceeded the Maximum Microbial Contamination Limit (BMCM).

Based on the results of the t-test, it was found that the number of *Escherichia coli* bacteria in chicken slaughterhouses and traditional markets was not significantly different ($P>0.05$), so there was no significant difference between the average number of Escherichia coli contamination in samples of broiler chicken sold in chicken slaughterhouses and traditional markets in the city of Medan. The average at the slaughterhouse was 3.15 log CFU/g, and the average at the traditional market was 3.39 log CFU/g.

According to Gustiani [12] and [13] that contamination of meat by microbes can occur before and after the animal is slaughtered. Sources of contamination can come from animals (skin, nails, offal), workers/humans who can contaminate livestock products, equipment (knives, cutting tools/cutting boards, boxes), buildings including floors, and the environment (air, water, soil), and packaging.

Bacterial Analysis *Salmonella sp.*

The analysis of *Salmonella sp.* was conducted on samples of broiler chicken marketed at chicken slaughterhouses and traditional markets in Medan. This examination is based on the conditions set out in SNI 3924:2008; it is stipulated that chicken meat may not contain Salmonella (negative/25 grams). The results of Salmonella sp bacteria examination on 30 samples of broiler chicken meat at chicken slaughterhouses and 30 broiler chicken meat at traditional markets can be seen in Table 7 below.

Table 7. *Salmonella sp.* on broiler chicken meat at chicken slaughterhouses and traditional markets.

Origin sample	amount	Contamination		Total
		Positive	Negative	
Slaughter	30	17	13	17/ 30
Traditional marks	30	20	10	20/30

Note:[tn]= chi-square test results were not significantly different

The analysis based on Table 7 shows that the research results on 30 samples of chicken meat at the chicken slaughterhouse found 17 samples contaminated with *Salmonella sp*. Analysis of *Salmonella sp.* laboratory tests showed that 17 out of 30 (56%) samples of chicken meat at the slaughterhouse contained Salmonella sp. and the percentage that 20 out of 30 (66%) samples of chicken meat in traditional markets contain Salmonella sp. According to research by [8] that samples of chicken meat from two different locations obtained a percentage of 55%, namely 17 out of 40 samples (42.5%) from traditional markets, while 5 out of 40 samples (12.5%) from supermarkets)

Based on the results of the Chi-square test, the calculated χ2 value $\leq \chi^2$ table and the level of *Salmonella sp.* in samples of chicken meat from chicken slaughterhouses was 56%, while the level of S*almonella sp.* in samples of broiler chicken meat originating from traditional markets is 66%, that the frequency of observations with the expected frequency of samples at chicken

slaughterhouses and traditional markets are not significant differences that both chicken slaughterhouses and traditional markets are polluted with *Salmonella sp*. Although there are some differences in sanitary hygiene in chicken slaughterhouses and traditional markets, it cannot rule out the possibility that the chicken meat is not contaminated with Salmonella sp. bacteria.

Microbial contamination in poultry products can also be caused by livestock manure that has been infected with Salmonella, which contaminates the feed and drinking areas of livestock so that it enters and multiplies in the digestive tract of poultry. Another thing that might happen is when cutting and packing with a knife or other tools; the carcass is contaminated with Salmonella bacteria more after the slaughter process than before.

4   Conclusions

Contamination of *Escherichia coli* in samples of broiler meat originating from chicken slaughterhouses and traditional markets in Medan city can be stated that all of them exceed the threshold determined by SNI 3932: 2008, namely 1×10^1 CFU/g, while the level of contamination of *Salmonella sp.* in the sample of chicken meat that comes from chicken slaughterhouses is 56% and that comes from traditional markets is 66%.

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