Detection of coliform bacteria, total plate count and pH value in chicken eggs from Maros traditional market

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Abstract. Indonesian consumers generally buy chicken eggs in traditional markets. Mostly, the traditional markets were less clean places, making it easier for bacterial contamination to the commodity. The purpose of this study was to determine the presence of Coliform bacteria, Total Plate Count (TPC) and pH value of chicken eggs from the traders in traditional markets in Maros regency. Chicken egg samples were obtained from 3 traditional markets in Maros Regency, each from 3 sellers and 3 replications. The number of coliform bacteria was determined using Brilliance Agar media while the TPC was determined using Plate Count Agar. The pH value was measured using a pH meter. The research method used was a pour plate to count Coliform and TPC bacteria then analyzed descriptively and presented in tabular form. The pH value was analyzed using analysis of variance and further test of the least significant difference. Based on SNI-3926-2008 regarding the microbiological quality requirements of fresh eggs, all egg samples met the Coliform and TPC bacteria standard, with the number of $1.1 \times 10^1$ and $2.3 \times 10^4$, respectively. The pH of eggs was still in the normal range with an average of 7.1 although the type of market affects the pH of the eggs. All egg samples studied based on Coliform bacteria, TPC and pH were categorized as safe for consumption.

1. Introduction
Eggs are source of animal protein, highly nutritious, palatable, easy to obtain, affordable prices, and can be made various products such as bread, cakes, salted eggs and others. The nutritional composition of eggs consists of 73.7% water, 12.9% protein, 11% fat and 0.9% carbohydrates. Almost no fat content in egg whites, the fat in eggs is found in egg yolks, which reaches 32%, while egg whites contain very little fat [1].

According to Afiyah and Rahmawati (2017), the supply chain of eggs used a delivery system. In the supply chain from breeders to retailers, eggs were collected on egg racks under varies conditions [2]. Egg storage required a safe container, avoid from heat, in a warehouse or storage room. People in Indonesia generally buy chicken eggs in traditional markets. Mostly, the traditional markets were less clean places, making it easier for bacterial contamination. The dirty and less hygienic place enabled microbial contamination to the egg, hence may harm the consumers. One of the contaminant bacteria is Coliform. The presence of Coliform in food shows the possibility of microbes that are toxic to health. Disorders caused to
consumers are nausea, abdominal pain, vomiting, diarrhea, bloody bowel movements, high fever, even in some cases seizures and lack of fluids or dehydration [3].

Apart from microbial contamination, another thing that needs to be considered is the physical quality of food, one of which is pH. pH is one of the factors determining the internal quality of eggs. This occurs due to the evaporation of a lot of liquid and gas in the eggs, causing a lot of the internal quality of the eggs to have decreased when they are consumed by consumers. The longer the storage time, the greater the evaporation of liquids and gases in the eggs, so that it will cause a bigger air cavity which causes the egg white to become thick and thin [4].

2. Methods

2.1. Sampling methods
Chicken egg samples obtained from three traditional markets in Maros regency. From 9 sellers, a total of 135 chicken eggs were collected then grouped into 27 samples (1 sample consisted of 5 chicken eggs). Each sample was put in a sterile plastic bag, labeled and transported to the Veterinary Public Health Laboratory of the Disease Investigation Center, Maros, for bacteria and pH testing.

2.2. Bacteria enumeration method
Coliform count and total plate count were performed using serial dilution and pour plate methods. Dilution was carried out using buffered peptone water (BPW) with a sample ratio to BPW of 1: 9. The egg samples and BPW were homogenized using stomacher at 230 rpm for 2 minutes. The Coliform plating was carried out using Brilliance Agar media and incubation was carried out at 37°C for 24 hours. Coliform bacteria were shown with pink colonies on Brilliance Agar media. Plating for TPC calculation is done using Plate Count Agar (PCA) media and incubation was carried out at the temperature of 34–36°C for 48 hours. The number of bacteria was calculated using the following formula [5].

\[ \text{Bacteria number} = \frac{\text{colony number}}{\text{dilution factor}} \]

2.3. pH value determination
The pH measurement on chicken eggs was carried out using a pH meter. The pH meter is neutralized by dipping the pH meter stick into the aquadest solution then dipping the stick into the sample solution. Measurements were carried out 3 times in each sample, then the results were averaged [6].

2.4. Data analysis
Result data on the number of Coliform bacteria and TPC were analyzed descriptively and presented in tabular form. The pH value data were analyzed using analysis of variance, then further testing using least significance difference [7].

3. Results and discussion
Apart from physical and chemical aspects, microbiological quality also determines the safety and suitability of a food material for consumption. The higher the level of Coliform bacteria contamination, the higher the risk of the presence of other pathogenic bacteria. The presence of Coliform is an indication of inadequate sanitary conditions. Hygiene and sanitation is an effort to reduce or eliminate the factors that cause pollution of the facilities used for the storage and marketing processes [8]. Environmental sanitation also affects the presence of Coliform bacteria contamination. The Coliform and TPC enumeration of chicken egg samples results were presented in table 1.
Table 1. The average number of Coliform and TPC of chicken egg samples from Maros traditional market

| Market | Coliform (CFU/g) | TPC (CFU/g) |
|--------|-----------------|-------------|
| A      | 1.1×10¹         | 2.8×10⁴     |
| B      | 1.1×10¹         | 4.0×10⁴     |
| C      | 1.1×10¹         | 1.1×10³     |
| SNI    | 1.0×10²         | 1.0×10⁵     |

SNI-3926-2008 regarding the microbiological quality requirements of fresh eggs.

The results in table 1 showed that the average number of Coliform in market A, B, and C was 1.1 x 10¹. The Coliform number met the SNI-3926-2008 [9] regarding the microbiological quality requirements of fresh eggs, as all the egg samples were safe for consumption. Coliform bacteria, if found in fresh eggs, is an indicator of contamination due to poor handling of eggs. Contamination of eggs can occur vertically and horizontally. Vertical or transovarial contamination is originating from infected hens, while horizontal contamination occurs outside the hen. Horizontal contamination occurs when bacteria enter eggs, for example, from dirt that sticks to the egg shells such as dust, soil and feces [10].

The TPC enumeration test was carried out to determine the number of bacteria contained in eggs by counting bacterial colonies. Result in table 1 showed that the TPC in all egg samples were below the SNI-3926-2008 requirement, as of safe for consumption. The highest increase in bacterial numbers occurred in market storage [11]. Dirt will stick to the shell and over time will enter the egg through the pores. Humans are one of the factors in the occurrence of contamination, when sellers or buyers repeatedly hold eggs from one egg to another that contain chicken droppings.

Table 2. The pH value of chicken egg samples from Maros traditional market

| Market | pH value |
|--------|----------|
| A      | 7.4c     |
| B      | 6.9a     |
| C      | 7.1b     |
| Average| 7.1      |

abc different superscript showed highly significance difference (p<0.01)

High temperature stimulates the growth of the extrinsic and intrinsic microbes in the contents of the eggs. High temperature also accelerates weight loss due to water evaporation or dilution of egg contents, thus the air space in the eggs enlarges due to evaporation, and increases in pH due to CO₂ loss [12]. In addition, microbes that stick to the surface of the egg shell will enter the egg through the pores in the eggshell. The breakdown of proteins by microbes causes the formation of ammonia which can increase the pH value. The average pH value of chicken egg samples obtained from three traditional markets in Maros was presented in table 2. The result showed that the pH value of eggs was still in the normal range with an average of 7.1 although the type of market affects the pH of the eggs. From interview with the sellers, the egg samples...
obtained from 3 traditional markets in Maros were still newly laid, which was less than 1 week of ovoposition.

Animal origin food are perishable products, thus maintaining the food safety and quality is very important. The food safety assurance is obtained through the application of a food safety system in each production process, such as the good farming practices (GFC) system, the good manufacturing practices (GMP) system that is applied during processing, good handling practices (GHP) is a system applied when handling the post-harvest stage, good distribution practices (GDP) is a system applied when distributing products to consumers [13]. The high temperature prevents the eggs from surviving long after ovoposition. The factors that influence the decline in egg quality were shelf life, temperature and relative humidity during storage [4]. According to SNI-3926-2008 [9], a proper eggs storage condition is maximum of 14 days at room temperature with a humidity between 80% and 90%, or maximum of 30 days at temperatures between 4°C and 7°C with a humidity between 60% and 70%.

4. Conclusion

Based on SNI-3926-2008 [9] regarding the microbiological quality requirements of fresh eggs, all egg samples obtained from traditional markets in Maros, met the Coliform and TPC bacteria standard, with the number of $1.1 \times 10^1$ and $2.3 \times 10^4$, respectively. The pH of eggs was still in the normal range with an average of 7.1 although the type of market affects the pH of the eggs. All egg samples studied based on Coliform bacteria, TPC and pH were categorized as safe for consumption.

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