

ANALYSIS OF ESCHERICHIA COLI AND SALMONELLA SP. CONTAMINATION IN DRINK SNACKS SOYBEAN MILK IN PADANG

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Abstract

Soybean is a food that has many benefits. One of the benefits is as an ingredient in making soy milk drinks. This drink is made with finely ground soybeans, added with water and filtered, which then produces a white liquid that resembles milk. But if the manufacturing process is not good and clean, it can be contaminated by several pathogenic bacteria such as; Escherichia coli, Salmonella sp., Staphylococcus aureus, and Bacillus cereus. The existence of these pathogenic bacteria to food products can cause various diseases in people who consume these products. Therefore the safety of a food product consumed can be determined from the amount of pathogenic bacteria contained in a food product. Based on this, this study aims to analyze and find out the contamination of pathogenic bacteria Escherichia coli, Salmonella sp., Which is found in soy milk snacks in Padang City. This research is a type of descriptive research using a qualitative approach in the form of observation, interviews and laboratory tests. Samples were taken by purposive sampling method, as many as 9 samples in several markets in Padang City. The results showed that the samples tested positive for Escherichia coli, Salmonella sp., and had a higher value than the maximum threshold required in SNI.

Keywords: *Escherichia coli, Salmonella sp, Soybean Milk*

INTRODUCTION

Soy milk is one of the food products that have several advantages, including being cheaper than cow's milk, high nutritional value, suitable for patients with lactose intolerance and does not cause allergies for consumers. Soy milk protein has an amino acid structure that is almost the same as cow's milk, so soy milk is often used as a substitute for cow's milk. Besides, this drink also contains fat, carbohydrates, phosphorus, iron, provitamin A, vitamin B complex except for B12 (Singh, 2009).

Public awareness about the cleanliness of soy milk is something that needs attention because food or drinks that contain contaminated ingredients when consumed will cause foodborne illness. Foodborne illness from bacteria can be in the form of intoxication or infection. Detoxification is caused by the presence of bacterial toxins that form in food when bacteria multiply, while the infection is caused by the

entry of bacteria into the body through contaminated food and the body responds to these bacteria. Bacteria that cause this disease are known as pathogenic bacteria.

Food and drinks that are contaminated by bacteria are caused by eating foods that are still raw, contaminated by feces, milk pasteurized soft cheese, and foods that are processed using water that has been contaminated by bacteria (Andriani, 2005). Most human diseases are caused by water that has been contaminated by pathogenic bacteria such as diarrhea, typhus, dysentery, botulism, and hepatitis A (Winarno 1997). Bacteria that cause the disease include *Escherichia Coli*, *Listeria*, *Campylobacter*, *Salmonella sp. sp.*, *Staphylococcus aureus*, and *Bacillus cereus* (Andriani, 2005; Supar 2005).

Several studies have made observations on the presence of microbial forms in food and drinks sold in several regions. Sirait's research (2009) in Medan City, showed that in small businesses processing soy milk does not meet the health requirements of a beverage. In Yogyakarta using the MPN method found that in 21 dawet ice samples, 100% contained coliform which could be an indicator of the presence of *Escherichia Coli* (Fatimah, 2017). Another study conducted by Sari (2009) on green grass jelly drinks sold in Pasar Raya Kota Padang, also found that all samples tested positive for *Escherichia Coli* bacteria ranging from 96 to 240 in 100 ml samples. Plus in 2016 the city of Padang is included in the category of the five largest number of cases of diarrheal disease in infants from all districts/cities in West Sumatra. (West Sumatra Province Health Service, 2016). Therefore the dangers caused by these microbes need to be analyzed and dealt with further so that the case of food poisoning is not widespread and is detrimental to public health.

Based on preliminary observations in several markets in the city of Padang showed that the storage of soy milk drinks are not stored in a cooling place, sell in places where the crowd and open, and the presentation using plastic cups or plastic bags that are not guaranteed clean. This results in a high chance of pathogenic bacterial contamination. Based on this, the writer wants to know the quality of soy milk in Padang, especially the bacterial content in it, by using the standards set by SNI.

METHOD

This type of research is an observational descriptive with a purposive sampling technique and obtained 9 samples of soy milk drinks sold in large markets in the city of Padang. Sample testing was carried out using the Total Plate Count (TPC) and Most Probable Number (MPN) methods.

The target population in this study are traders who sell soy milk snacks at several large markets in the city of Padang (Figure 1) between the markets in Kec. Koto Tengah Lubuk Buaya, Kec. Padang Timur Simpang Haru, Kec. Lubuk Kilangan Bandar Buat, Kec. Lubuk Begalung Parak Laweh, Ke. Nanggalo Pasar Siteba, Kec. Pauh Pasar Baru, Alai Market, Kec. Padang Barat Purus, Kec. Kuranji Pasar Belimbing. The object of this research is soy milk snacks in Padang City which is sold using plastic packaging. The research sample was taken by means of purposive sampling that is choosing a sample with the criteria set by researchers



Figure 1. Samples of soy milk drinks

The measurement aspect of this study is the number of pathogenic bacteria in soy milk drinks sold in several markets in the city of Padang. Measurements made by laboratory examinations are supported by observations and interviews as supporting data. The laboratory results were then matched with SNI 01-3830-1995, fulfilling the requirements if the total E. coli bacteria <3 MPN / ml, Salmonella sp. sp must be negative.

Sampling was conducted at sellers of soy milk snacks at a large market in the city of Padang. The Total Plate Count (TPC) and Most Probable Number (MPN) were carried out at the Agricultural Microbiology and Biotechnology Laboratory of Andalas University. Samples are taken and stored in a cool box and tested directly in the laboratory.

The tools used in this study were analytical scales, autoclaves, incubators, micro pipets, blunts, vortexes, bunsen, colony counters, laminar airflow, test tube racks, Durham tubes, measuring pipettes, pencils, cotton, rubber rubbers, sterile spoons, sterile tubes, reaction, Petri dish dish, Erlenmeyer, hotplate, glass beaker, test tube, aluminum foil, label. The materials used in this study were soy milk drinks, TSA (Tryptone Soya Agar), SSA (Salmonella sp. Shigella Agar), PCA (Plate Count Agar), LBDS (Lactose Broth Double Strength), LBBS (Lactose Broth Single Strength), MSA (Mannitol Salt Agar), Physiological Salt, sterile distilled water, 70% alcohol.

The data obtained were analyzed using simple statistics with a descriptive approach. The data that has been processed is then assessed the quantity and quality of the presence or absence of pathogenic bacteria (Escherichia Coli and Salmonella sp. Sp.). The steps in this research can be seen in the flow of research activities that can be seen in Figure 2.

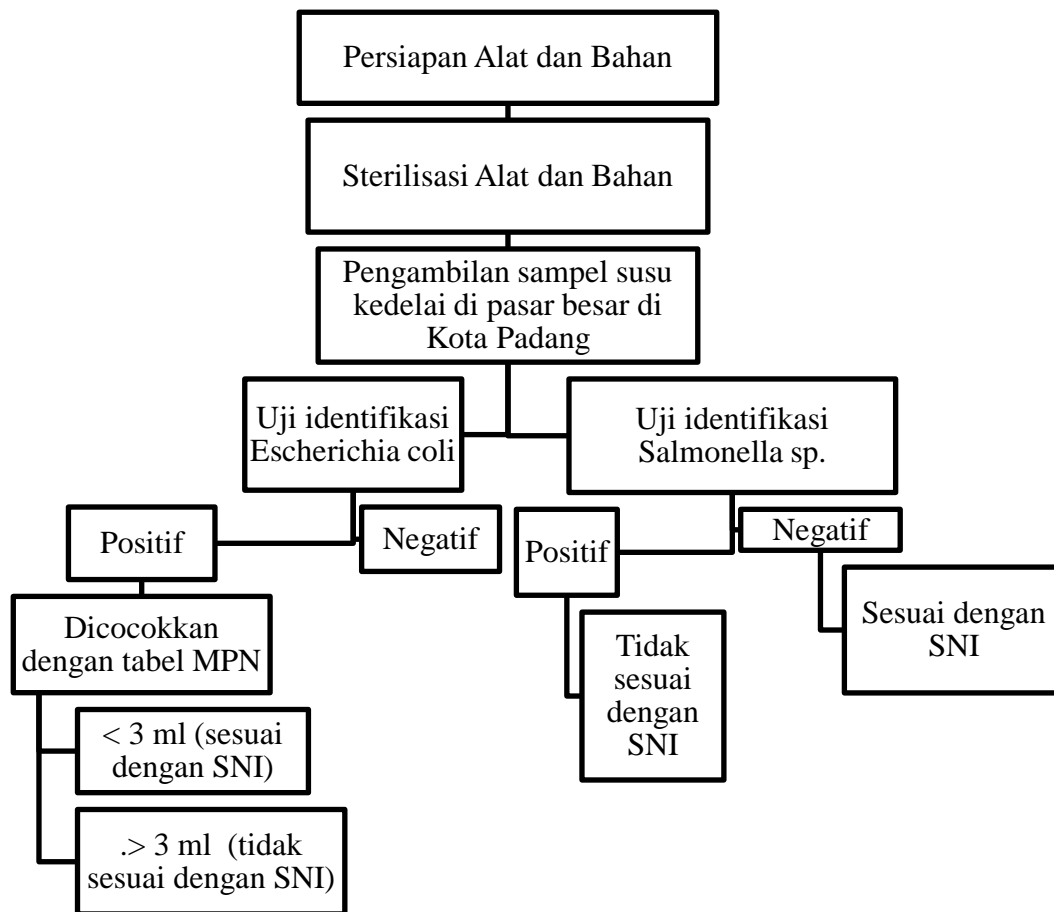


Figure 2. Picture Flow of Research Activities

RESULT AND DISCUSSION

Number of Escherichia Coli bacteria in soy milk snacks in Padang City

The results of the calculation of the number of pathogenic bacteria Escherichia Coli in soy milk snacks in Padang City can be seen in Table 3 below. Based on Table 3 below, it can be seen that from the samples of soy milk drinks tested there were positive samples containing pathogenic bacteria Escherichia Coli and the amount exceeds the limit value required by SNI. From 9 samples that were tested by Duplo repetition, it was found that most of the samples tested positive contained Escherichia Coli bacteria and the amount was also greater than the limit value of soy milk beverage quality requirements according to SNI 01-3830-1995.

Table 1 Jumlah Bakteri *Escherichia Coli* pada minuman jajanan susu kedelai di Kota Padang

| SAMPLE CODE | INDEKS APM/ML | APM/ML SESUAI SNI | INFORMATION |
|-------------|---------------|-------------------|------------------|
| KTLB1 | 210 | | TIDAK SESUAI SNI |
| KTLB2 | 64 | | TIDAK SESUAI SNI |
| PTSH1 | 0 | | SESUAI SNI |
| PTSH2 | 9 | | TIDAK SESUAI SNI |
| LKBB1 | >1100 | | TIDAK SESUAI SNI |
| LKBB2 | >1100 | | TIDAK SESUAI SNI |
| LBPL1 | 23 | | TIDAK SESUAI SNI |
| LBPL2 | 240 | | TIDAK SESUAI SNI |
| NS1 | 3 | <3 | SESUAI SNI |
| NS2 | 9 | | TIDAK SESUAI SNI |
| PPB1 | 0 | | SESUAI SNI |
| PPB2 | >1100 | | TIDAK SESUAI SNI |
| PA1 | 120 | | TIDAK SESUAI SNI |
| PA2 | 0 | | SESUAI SNI |
| PBP1 | 3 | | SESUAI SNI |
| PBP2 | 0 | | SESUAI SNI |
| KB1 | 4 | | TIDAK SESUAI SNI |
| KB2 | 0 | | SESUAI SNI |

The results of the test showed that the majority of positive samples formed bubbles / gas, which was suspected to have been contaminated by *Escherichia Coli* bacteria (Figure 3). Kusuma (2009) describes the process of sugar (lactose) fermentation in LB (Lactose Broth) media due to the presence of fecal coliform bacteria (*Escherichia Coli*). Sugar fermentation in the presence of energy produced by bacteria will produce pyruvic acid and acetic acid, then CO₂ gas bubbles appear in the media. The test tube is tightly closed, causing carbon gas will push the space in the Durham tube. If in more than 24 hours, more gas space will be formed in the Durham tube in a positive reaction. The negative reaction does not indicate the presence of bacteria characterized by the absence of gas bubbles in the Durham tube. The formation of bubbles/gas and color changes indicate the occurrence of lactose fermentation in liquid lactose media by bacteria present in soy milk, so as to produce acids and gases.

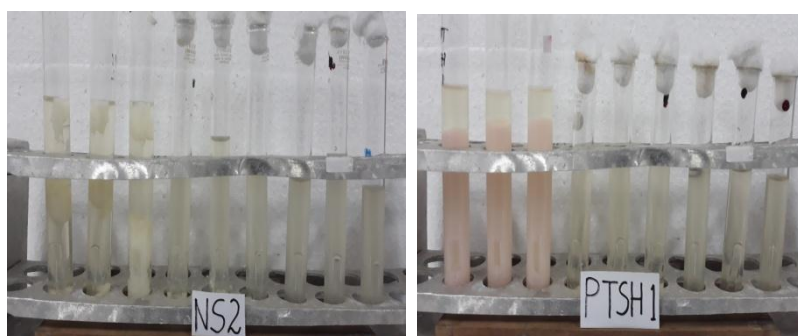


Figure 3. Bubbles / gas and discoloration in the durham tube on the test of the existence of *Escherichia Coli*

The presence of gas bubbles in the test results can be interpreted that soy milk beverage samples are not suitable for consumption because they contain pathogenic bacterial contamination whose amount exceeds the requirements set by SNI that is <3 ml. The presence of these pathogenic bacteria will have an impact on health problems when the product is consumed. The presence of Escherichia Coli bacteria in the product can be caused by several things, namely: the quality of the water used in the production process, the hygiene and sanitation of people who process the products and equipment used, as well as the sanitation of the environment around the processing site (Kusuma, et al., 2016).

The quality of water used during the processing of soy milk greatly affects the quality of soy milk produced later. The use of unclean water is a gap for breeding grounds for bacteria and viruses that cause disease, one of which is diarrhea. Water must be boiled until the pathogenic microbes die (Sumolang et al., 2019). According to Sumolang, et al. (2019) family environment is one of the factors that also supports efforts to maintain individual health. Several factors in the family environment associated with the incidence of diarrhea are the source, processing, and storage of drinking water. The source of drinking water is related to the incidence of diarrhea. Diarrhea is most common among people who use water from springs. Drinking water storage is also related to the incidence of diarrhea because storing water in open places is very vulnerable to the occurrence of pathogenic microbial contamination that causes disease. In addition to water quality factors, individual hygiene and sanitation (food processors and handlers) will also affect product quality.

Pathogenic bacterial contamination can occur due to the behavior of not washing hands when hands are dirty and after urinating or defecating. Washing hands is an important activity because hands will come into direct contact with food. The processing of food which has been hygienic but when touched with dirty hands causes food to be contaminated by pathogenic bacteria which can cause disease including diarrhea. Food processors and food handlers must clean their hands using clean water and use soap and nail brushes before touching food, handling food utensils, before eating, after leaving the toilet or toilet and after removing the stool.

The number of Salmonella Bacteria sp. in soymilk snacks in Padang City

The results of counting the number of pathogenic bacteria Salmonella sp. in soy milk snacks in Padang City can be seen in Table 4 below.

Based on Table 4 it can be seen that the results of laboratory tests of soy milk beverage samples obtained the results that from 9 samples tested by a repetition of Duplo obtained results that most of the positive samples contain pathogenic bacteria Salmonella sp. sp. (Figure 4). This is not following the quality requirements of soy milk drinks stipulated in SNI 01-3830-1995. According to SNI 01-3830-1995 regarding the quality requirements of soy milk drinks, the presence of Salmonella sp. sp must be negative in soy milk drinks. The results of this test indicate that a positive soy milk beverage product contains Salmonella sp. sp. unfit and may not be consumed because it will cause disease.

Table 2 Number of Salmonella sp. Bacteria. sp. in soy milk drinks in the city of Padang

| SAMPLE CODE | AMOUNT OF COLONIES | INFORMATION | AMOUNT OF COLONIES ACCORDING TO SNI | SNI CRITERIA |
|-------------|--------------------------------------|-------------|-------------------------------------|------------------|
| KTLB1 | $<3,0 \times 10^2$ | POSITIVE | | TIDAK SESUAI SNI |
| KTLB2 | 0 | NEGATIVE | | SESUAI SNI |
| PTSH1 | 0 | NEGATIVE | | SESUAI SNI |
| PTSH2 | 0 | NEGATIVE | | SESUAI SNI |
| LKBB1 | $<3,0 \times 10^3$ | POSITIVE | | TIDAK SESUAI SNI |
| LKBB2 | $2,0 \times 10^3$ | POSITIVE | | TIDAK SESUAI SNI |
| LBPL1 | 0 | NEGATIVE | | SESUAI SNI |
| LBPL2 | 0 | NEGATIVE | | SESUAI SNI |
| NS1 | $<3,0 \times 10^1 (1,0 \times 10^1)$ | POSITIVE | NEGATIF | TIDAK SESUAI SNI |
| NS2 | $3,3 \times 10^2$ | POSITIVE | | TIDAK SESUAI SNI |
| PPB1 | 0 | NEGATIVE | | SESUAI SNI |
| PPB2 | 0 | NEGATIVE | | SESUAI SNI |
| PA1 | $<3,0 \times 10^1 (1,2 \times 10^2)$ | POSITIVE | | TIDAK SESUAI SNI |
| PA2 | 0 | NEGATIVE | | SESUAI SNI |
| PBP1 | $<3,0 \times 10^1 (1,0 \times 10^1)$ | POSITIVE | | TIDAK SESUAI SNI |
| PBP2 | 0 | NEGATIVE | | SESUAI SNI |
| KB1 | $3,0 \times 10^3$ | POSITIVE | | TIDAK SESUAI SNI |
| KB2 | 0 | NEGATIVE | | SESUAI SNI |

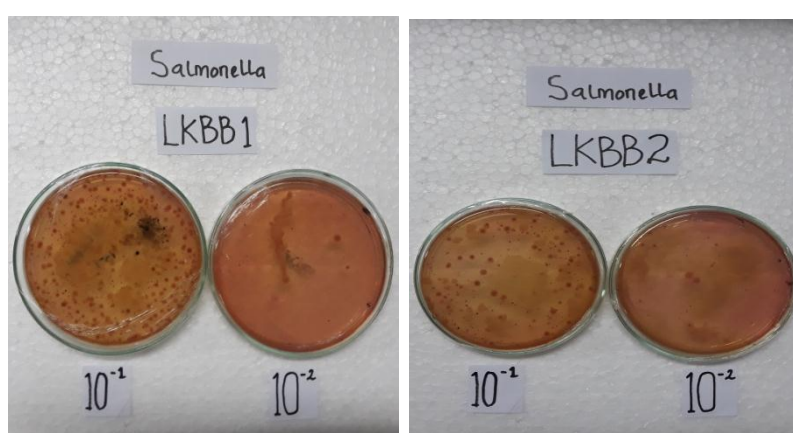


Figure 4. Positive samples containing *Salmonella* sp.

According to Khaq, et al. (2016), Salmonella sp. sp. can be caused due to imperfect soybean seeds soaking during the processing, soaking the soybean seeds will cause a decrease in pH to become acidic (5 to 4) due to the fermentation process by lactic acid bacteria. Contaminant bacteria such as Salmonella sp. sp. cannot grow with a low pH (acid). However, if the pH cannot reach the acid due to the lack of immersion, it will trigger the growth of contaminants.

Presentation and Storage of Soy Milk Drinks

the product is not stored in the cooler. In selling their wares, most traders place their products in the open air which can be directly exposed to hot air. With this selling technique, the risk of contaminating the product with contaminant bakery will increase, especially if they sell it for a long time, from morning to evening. In her research Putri, et al., (2018) stated that the length of the period used in food handling since the food was finished cooking, packaging until distribution, is the main cause of food poisoning. Food contamination occurs due to bacteria, air, food handlers, and equipment used. A long-time accompanied by a good storage process can trigger the growth of contaminant microbes well.

CONCLUSION

Based on the results of research that has been done, the contamination of pathogenic microbes in soy milk snacks in Padang City is still high and these results indicate that soy milk drinks are not suitable for consumption. The results of pathogenic microbial testing Escherichia Coli and Salmonella sp. sp. in soy milk snacks still exceed the safe limits set by SNI 01-3830-1995 concerning the quality requirements of soy milk drinks.

In further research it is recommended to conduct an analysis of the quality of soy milk drinks based on nutritional values referring to SNI 01-3830-1995 concerning the quality requirements of soy milk drinks so that the quality of soy milk drinks sold in the city of Padang can be controlled and truly guaranteed good quality nutritional quality and microbiological quality.

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