

RESEARCH STUDY

English Version

OPEN ACCESS

Hygiene Sanitation and Escherichia Coli Bacteria on Clover Leaves as Surabaya Traditional Culinary (Study in Kampoeng X, Y District, Surabaya)

Hygiene Sanitasi dan Keberadaan Bakteri Escherichia Coli pada Semanggi sebagai Jajanan Tradisional Surabaya (Studi Di Kampoeng X, Kecamatan Y, Surabaya)

Anita Dewi Moelyaningrum^{1*}, Resty Ayu Permatasari¹, Ninna Rohmawati²¹Department of Environmental Health, Public Health Faculty, Universitas Jember, Jember, Indonesia²Department of Nutrition, Public Health Faculty, Universitas Jember, Jember, Indonesia**ARTICLE INFO**

Received: 12-09-2022

Accepted: 06-03-2023

Published online: 05-09-2023

***Correspondent:**

Anita Dewi Moelyaningrum

anitadm@unej.ac.id

DOI:

10.20473/amnt.v7i3.2023.390-399

Available online at:<https://e-journal.unair.ac.id/AMNT>**Keywords:**

Hygiene, Sanitation, Clover Leaves, E. Coli

ABSTRACT

Backgrounds: Clover food, or Semantic, is one of the traditional Surabaya culinary delights and is still the most favorable for various communities and tourists for its unique taste. Clover food is one of the potential foods to be contaminated by microbes because it does not implement hygiene and sanitation to handle it.

Objectives: This research was conducted to analyze the hygiene sanitation and the Escherichia coli bacteria at clover food Surabaya, Indonesia

Methods: This type of research was descriptive with Interviews, observation, and laboratory tests collected data. The total samples examined were 32 respondents and 64 clover samples consisting of 32 clovers and 32 seasonings.

Results: Almost all of the respondents had a higher level of knowledge (93.8%), had applied personal hygiene in the excellent category (59.4%), and had applied sanitation of food processing that was well-rated (87.5%). According to the lab, there were two positive samples of E. Coli.

Conclusions: Almost all respondents had not applied appropriate personal hygiene, and E. Coli bacteria were still found in the clover food. One was found in highly-educated respondents who applied food processing sanitation well.

INTRODUCTION

Food can be a medium for disease transmission¹. Food-related diseases are often caused by microbiological contaminants such as bacteria, viruses, parasites, or other toxic substances in food². Food that enters the body must pay attention to aspects of good hygiene and sanitation³. According to the Ministry of Health of the Republic of Indonesia in 2004⁴, so that poisoning and transmission of a disease do not occur, it is necessary to implement food sanitation hygiene measures, including traditional food^{5,6,7,8,9}. There are six principles of food sanitation hygiene, namely, selection of food raw materials, storage of raw materials, food processing, food storage, food transportation, and food serving. The behavior of workers or food handlers also plays a role in determining the product quality to be produced¹⁰. So the health and hygiene of workers or food handlers also need attention.

Foodborne diseases are diseases caused by consuming contaminated food. Food contamination can be caused by microbes such as Escherichia coli bacteria. E. Coli can be used as an indicator of food contamination,

which causes foodborne disease¹¹. According to the Regulation of the Minister of Health of the Republic of Indonesia No. 1096 of 2011, Escherichia coli bacteria should not be present in food (0/gram)¹². E. Coli can come from various sources, including the water used to process food¹³. According to the 2019 Food and Drug Supervisory Agency (BPOM) annual report¹⁴, 53 cases of food poisoning extraordinary events (KLB) were reported with the principal causative agent, microbiological agents, 35 incidents, with 16% originating from snacks. In 2017-2019, food poisoning outbreaks in Surabaya increased in 2018¹⁵ and 2019¹⁶. Profile data from the Surabaya City Health Office in 2016, the total number of cases of diarrhea in Benowo District found 1,552 cases of diarrhea. The foodborne disease often causes digestive disorders such as diarrhea, which are hygiene and sanitation conditions related to diarrhea¹⁷. Of the total cases of diarrhea, 1,125 cases (72.48%) were handled by the Sememi Health Center Surabaya¹⁸.

Semanggi snacks are one of the traditional snacks from the city of Surabaya, which are in great demand by various groups of people and are hunted by

many tourists because of their unique taste. *Semanggi* hawker producer, localized in one location. "Kampoeng X" is a center for producers and traders of *Semanggi* snacks in Surabaya. *Semanggi* snacks only exist in Surabaya, so they are a culinary icon of Surabaya. However, *Semanggi* snacks are one of the foods that have the potential to be contaminated with microbes because it is served not hot, not in closed conditions, the selling place is on the side of the road, and sometimes, in preparing food, handlers do not use gloves or food tongs. Although *Semanggi* snacks have never been the cause of food poisoning outbreaks, the high potential for contamination must be prevented. As a culinary icon of Surabaya, efforts are needed to maintain the hygiene and sanitation aspects of food so that traditional *Semanggi* snacks can still be enjoyed by the public and tourists safely. This study aimed to describe sanitary hygiene and identify the presence of *E. Coli* bacteria in *Semanggi* snacks in Kampoeng X Surabaya.

METHODS

This study was descriptive research. Data was collected by interviews, observation, documentation, and laboratory tests to determine the presence of *E. Coli* bacteria in ripe clover leaves and spices. The presence of *E. Coli* was confirmed by the Isolation Identification method of *E. Coli*. This research was conducted in Kampoeng X, District Y, Surabaya, in September 2020. Village X is a center for producers and sellers of *Semanggi* snacks. All clover sold in the city of Surabaya comes from village X.

The population in this study were all clover traders from village X, totaling 45 people. All clover traders are also producers. They make clover snacks as well as market clover snacks throughout the city of Surabaya. Using the Slovin formula calculations, a sample of 32 people was obtained, and respondents were to be interviewed and observed. Each trader took clover samples (32 samples) and clover seasoning samples (32 samples) to take 64 samples. The research instrument in the form of interviews and observation sheets was made based on the instruments in the Minister of Health Regulation 1096/2011. Interviews were conducted to obtain data on the characteristics and knowledge of the respondents. Data on personal hygiene (14 questions) were obtained from interviews and observations, while sanitation data (47 questions) was obtained by observation. The presence of *E. Coli* was categorized into 2: positive *E. Coli* and negative *E. Coli* (Decree of the Minister of Health of the Republic of Indonesia No. 715 of 2003)

Sample examination was carried out using the isolation technique method, which was carried out in the Surabaya Health Laboratory Center (BBLKS). This research was approved by the Health Research Ethics Commission (KEPK) Faculty of Dentistry, University of Jember No.982/UN25.8/KEPK/DL/2020. Data processing techniques included editing, coding, tabulating, and cleaning. Data analysis used SPSS 22 software. The data was then presented descriptively using tables and cross-tabulations and translated into text or narrative. These data include the characteristics of the respondents (age, gender, last education, length of time selling, and nature

of service), level of knowledge of *Semanggi* snacks handlers (knowledge about efforts to avoid food contamination, knowledge of how to wash hands properly and correctly, knowledge of food hygiene factors, knowledge related to choosing a location and place of sale, knowledge about how to store food), how to make *Semanggi* snacks, application of personal hygiene food processing sanitation, and the presence of *E. Coli* bacteria on clover leaves and spices. Respondents' knowledge was grouped into three categories: good if they could answer ≥ 5 questions correctly, sufficient (2-3 questions), and poor if they only answered 1 question correctly.

RESULTS AND DISCUSSION

Characteristics of Respondents

Respondents in this study are *Semanggi* hawker traders who produce their wares independently and live in "Kampoeng X" Surabaya. This research was conducted in Kampoeng X District Y Surabaya with 32 respondents. The characteristics studied were based on age, gender, last education, length of time selling, and service. Age can affect the experience, knowledge, and insights of food handlers⁷. The results showed that the majority (43.8%) of respondents, namely *Semanggi* snacks handlers, were aged 40 - <60 years, and all were women. A person's level of maturity and strength in working and thinking will be more mature if the person is old enough. So, if we are old enough, the ability to carry out all activities related to thoughts, perceptions, and memories, and the ability to process information to gain knowledge and problem-solving abilities are included in the excellent category¹⁹.

Most respondents' last education (46.9%) graduated from elementary school, and 4 respondents (12.5%) did not attend school. Positive behavior can be influenced by the education one gets. So, the higher a person's education, the higher the awareness of the importance of maintaining hygiene and sanitation for *Semanggi* snacks will increase. As many as 53.1% of respondents claimed to have traded clover for over 5 years. Workers' skills in carrying out their duties can be affected by the length of service, where the more extended the period of service, the more it is expected that the behavior will be better²⁰. It is hoped that the period of selling can provide one's knowledge in maintaining food sanitation hygiene and positive behavior towards implementing it. There were 15 traders (46.9%) trading in the exact location (settled). Food handlers' knowledge regarding sanitation hygiene will be related to the quality of food served to consumers. Good knowledge will be able to produce good behavior in processing food so that contamination of food can be avoided. Most respondents (93.8%) have good knowledge regarding food sanitation hygiene. High knowledge about food sanitation hygiene procedures is expected to contribute to the behavior of respondents. Knowledge plays a vital role in the formation of behavior. The higher the knowledge that a person has, it can lead to perceptions or understandings, which in turn form attitudes that encourage the occurrence of a behavior. Complete data can be seen in Table 1.

Table 1. Frequency and distribution of respondents' characteristics related to sanitary hygiene and the presence of *Escherichia coli* bacteria in *Semanggi*

Characteristics of Respondents	Number of people (n)	Percentage (%)
Age		
Early Adults	12	37.5
Middle Ages	14	43.8
Elderly	6	18.8
Total	32	100
Gender		
Woman	32	100
Total	32	100
Last education		
No school	4	12.5
Graduated from elementary school/equivalent	15	46.9
Graduated from Middle School/Equivalent	4	12.5
Graduated from high school/equivalent	8	25
College	1	3.1
Total	32	100
Long Selling		
New (≤ 5 years)	15	46.9
Old (> 5 years)	17	53.1
Total	32	100
The Nature of Sales Services		
Stay	15	46.9
Semi Settled	9	28.1
Around	8	25
Total	32	100
Knowledge related to Sanitary Hygiene		
High	30	93.8
Moderate	2	6.3
Low	0	0
Total	32	100

Personal Hygiene of Respondent

The way of making clover snacks is done traditionally. The tools that must be prepared include a steamer (pot), basin, plate, spoon, filter, mortar, pestle, and banana leaf (for serving). The ingredients needed include clover, 3 g of sweet potato (yam), ½ kg of peanuts, ½ kg of brown sugar, 1 ounce of garlic, ¼ tablespoon of granulated sugar, 3 tablespoons of salt, and water. Vegetables for the main ingredients of clover snacks are clover leaves. The manufacturing process starts with washing the clover leaves and then drying them in the sun. The drying process is carried out two times. In the first drying, the clover leaves are dried in the sun for approximately six hours until the clover leaf stalks are completely dry. This step prevents clover leaves from becoming moldy when stored as stock during the rainy season. Clover plants are challenging to grow during the rainy season, and this is because the land used to plant clover is prone to flooding during the rainy season. At this stage, the clover is dried in the sun for six hours and then stored in a large plastic container. During the study, it was seen that clover leaves were dried in the sun in the yard and front of the house. Clover leaves that will be used immediately are then boiled and filtered using a filter.

After that, the clover leaves are squeezed using a cloth to remove the water content. The clover leaves that have been squeezed are then dried in the sun again. This second drying process is carried out for about 15 minutes so that the cloverleaf leaves do not stick between one leaf and the other and are not damp so they

do not rot quickly when carried around selling all day. Seasoning snacks starts with steaming the sweet potato for about an hour. Peanuts and garlic are fried and then pounded until smooth. Mix the steamed sweet potato with peanuts and garlic (already fried) and then grind again until slightly smooth. When sprinkled over the clover leaves, sprinkle the spices using boiled water. Next, steam the clover leaves on top of the pink-shaped banana leaves. Then, flush the cloverleaf with the seasoning, and the cloverleaf snacks are ready to be served.

The personal hygiene assessment in this study was based on the Decree of the Minister of Health of the Republic of Indonesia, Number 942 of 2003²¹, regarding guidelines for food sanitation hygiene requirements. The health of handlers is a requirement that must be met in processing food so that food does not become a medium for disease transmission from handlers to buyers²². The results of the observations showed that the condition of the respondents when in contact with food, all (100%) were not suffering from coughs, colds, influenza, or diarrhea during the study. This finding aligns with other research, which states that traders are in good health when selling²². All respondents covered their noses and mouths when cleaning or coughed while selling *Semanggi* snacks because, during the COVID-19 pandemic, food handlers always wore masks while selling clover. Masks can also prevent contamination when handlers sneeze or cough in front of processed food²³.

Food handlers should wear aprons and head coverings to minimize contamination from handlers into the food being processed. However, based on research, all respondents (100%) did not wear aprons or unique clothes when processing and serving clover snacks. Handlers come into direct contact with food, so they have to wear unique clothing for work or wear an apron. Dirt in work clothes has the potential to spread or fall into the food that is being processed so that food can become contaminated with bacteria²⁴.

All respondents did not wear head coverings when preparing *Semanggi* snacks at home. When selling, almost all respondents wore head coverings (veils), but during the processing process at home, the respondents did not wear head coverings, even though most of the processing of clover snacks is done at home. When selling at the location where the seller is selling, the respondent only carries out the presentation process to the buyer. Hair that falls in food is not the leading cause of bacterial contamination. However, this can reduce the aesthetic value of the food²⁴. In addition, all respondents (100%) admitted that they had scratched their limbs while working. Scratching the head or limbs should not be done when preparing food because bacteria in the body can enter the food.

There were 26 respondents (81.3%) who did not wash their hands using soap and running water. This does not follow the requirements in the Decree of the Minister of Health of the Republic of Indonesia no. 1096 of 2011, that both before and after processing food, workers must always wash their hands¹². Hand washing done correctly and adequately can remove dirt on the hands containing many bacteria or microbes. During the observation, it was also seen that there were respondents who always used hand sanitizer before serving customers or serving clover snacks. Washing hands using running water with soap or hand sanitizer is effective in reducing the number of microorganisms (anti-germ substances)²⁴. The results of observations that have been made show that as many as 15 respondents

(47.9%) wear dark clothes when preparing clover snacks. When working with food handlers, it is recommended to wear white or brightly colored clothes so that the clothes are easy to detect if they start to get dirty. Dirt in work clothes can spread or fall into the food being processed so that it can contaminate the food with bacteria²⁵. Six respondents (18.8%) used rings or bracelets during food processing. In making clover snacks, there are stages of washing, squeezing, and separating the clover leaves by hand. Suppose handlers do not remove their accessories or jewelry when in direct contact with food. The bacteria outside the skin under the ring or bracelet will contaminate the processed food. There were respondents (9.4%) whose fingernails were not long and dirty. Long nails get dirty and damp quickly, so between the fingers and nails, they can become a place for various dirt-containing microorganisms to stick. The bacteria nesting in the fingernails and between the dirty fingers can make the food that is processed contaminated²⁵. During observation, the respondents touched ripe clover leaves directly with their hands without using tools or gloves. Hands are one of the causes of the entry of contaminants into food. The hands move like scratching limbs, such as the nose, head, ears, and mouth, and holding sources of contaminants.

Of the 14 questions, the majority of respondents (59.4%) had personal hygiene, which was included in the excellent category (score 5-9 correct answers), and 13 respondents (40.6%) were included in the excellent category (Table 4). This finding is in line with research conducted on *Cenil* traders in the market⁷ and banana chips⁸, however, and it is different from the research conducted on *Pudak* sellers whose implementation of hygiene is still lacking²⁶. The application of personal hygiene handlers of *Semanggi* snacks is in a suitable category because some handlers still do not apply proper personal hygiene. All respondents did not wear aprons and head coverings when processing food. Most respondents also did not wash their hands before and after contact with food.

Table 2. Respondent's personal hygiene condition related to the presence of *Escherichia coli* bacteria in clover

Rating Items	Yes		No		Total	
	n	%	n	%	N	%
Not suffering from coughs, colds, influenza, diarrhea, infectious diseases such as typhoid, cholera, tuberculosis, hepatitis	32	100	0	0	32	100
Cover open wounds	32	100	0	0	32	100
Hands and fingers are clean	32	100	0	0	32	100
Routinely wash your hair at least twice a week when processing clover snacks	29	90.6	3	9.4	32	100
Fingernails are short and clean	29	90.6	3	9.4	32	100
Do not use jewelry or accessories (rings, bracelets, watches) when preparing clover food	26	81.3	6	18.8	32	100
Wear bright, bright-colored clothes (look clean)	17	53.1	15	47.9	32	100
Wearing an apron or unique clothes for work when preparing food from Clover	0	100	32	100	32	100
Wear a head covering when preparing clover food	0	100	32	100	32	100
The behavior of washing hands with soap and running water before and after handling food	6	18.8	26	81.3	32	100
Use of tools/equipment/hand mat	29	90.6	3	9.4	32	100
Do not smoke	32	100	0	0	32	100
Do not scratch limbs when preparing food	0	100	32	100	32	100

Rating Items	Yes		No		Total	
	n	%	n	%	N	%
Covering the nose and mouth when sneezing or coughing while preparing food	32	100	0	0	32	100

Hygiene and Sanitation in *Semanggi* Snacks Processing

Selection of Raw Materials

The interviews and observations show that all respondents (100%) chose suitable raw materials and fulfilled the requirements of the Regulation of the Minister of Health of the Republic of Indonesia No.1096 (2011)¹². Based on the results of interviews, cloverleaf leaves are used as ingredients for making clover snacks in a condition that looks clean and fresh (not wilted, not rotten, and color has not changed). Sweet potatoes used as seasoning ingredients are in good condition, such as hard skin (not mushy), smooth potato surface, not rotten, not sprouting, and not moldy. The peanuts used were also in good condition, not moldy or rotten. The brown sugar used is also not hollow or sandy; the surface looks flat and moldy, and the color is light to dark brown. As for the condition of the water used to grind the clover spices, the physical condition is colorless, odorless, and tasteless. Based on interviews, respondents admitted that they used refilled and boiled water (boiled themselves) to grind the spices.

Raw Material Storage

Storage of raw materials is storing food raw materials so that their quality is maintained. Most (78.1%) of the storage conditions for raw materials for clover snacks, such as sweet potato, clover leaf, peanuts, and brown sugar, are clean, well-maintained, and dust-free. Placement of raw materials (raw) Most of the respondents (96.9%) are also separated from prepared food. In line with the requirements of the Decree of the Minister of Health of the Republic of Indonesia No. 715 of 2003²⁷, the storage of raw and ready-to-eat food should not be combined in order to avoid cross-contamination. However, several respondents still attached raw materials to the floor (25%) and the wall (15.6%). This condition does not meet the requirements of Minister of Health Regulation No. 1096 of 2011¹², which is 15 cm between the food and the floor, 5 cm between the walls, and 60 cm from the ceiling so that the food does not come into direct contact with the floor, walls, or ceiling to prevent the storage area from becoming a nest for insects such as cockroaches or rats.

Food Processing

Food processing is converting raw materials or raw materials into clover snacks that are ready to be consumed. The kitchen owned by the respondent (50%) does not have ventilation. Activities in the kitchen produce higher temperatures, but it also produces hot steam and smoke. The presence of ventilation can make air circulation in the kitchen smooth²⁸. There were 30 respondents (93.8%) who had trash bins that did not meet the requirements in their kitchens. The trash bin owned by the respondent was only in the form of a plastic bag and was open. Owned trash does not have a cover. Trash without a cover will attract flies and other pests,

which then carry germs to contaminate food¹¹. Most of the kitchens (62.5%) do not have a ceiling. The ceiling has a function to prevent dust and other debris from falling, thereby minimizing contamination in processed food²⁹. None of the *Semanggi* snacks handlers dry the equipment using a dryer or clean cloth but only drain it on a rack or beside the dishwasher until it dries. This finding is in line with other studies, which state that, on average, respondents have washed equipment and then dried it by setting it on a rack and letting it dry on its own. If the equipment is not stored dry (still wet), dirt and dust will easily stick to and contaminate the equipment³⁰.

The source of water used for washing materials and tools came from PDAM water, namely, as many as 31 respondents (96.9%), while one respondent used well water, where the well is less than 10 meters from the septic tank and does not have a healthy lip. The construction of wells that do not meet technical requirements can be polluted. Water can be a medium for disease transmission. Bacteria, parasites, and viruses can move into the body through water. E. Coli bacteria in water indicates that feces have been contaminated. E. Coli is often found in water sources such as water^{13,31}. This can harm health because it has the potential to transmit disease. During observation, it was seen that the location where the clover leaves were dried was not free of dust because they were drying in front of the house on the village road, and there were still six respondents (18.8%) where the location where the clover leaves were dried contained insects such as flies. This could potentially make the clover leaves contaminated in the sun. This finding aligns with other studies showing that microbial contamination in food products can come from various sources. When food products are dried under direct sunlight in an open condition, they can cause contamination from the air. In addition, insects can land on food products dried in the sun³².

Finished Food Storage

The results showed that most respondents (90.6%) used containers (ripe clover leaves, seasonings, and other vegetable components) that were not completely closed, only covered with a napkin, plastic, or banana leaves. When serving customers, napkins, plastic, or banana leaves used as covers will be opened until the buying and selling process is complete. It is when they are open that clover leaves and spices have the potential to be infested with flies and exposed to dust because the majority of traders sell on the side of the main road. In addition, two food handlers (6.3%) still put cloverleaf with other vegetables such as sprouts and turi leaves together (not separately). Sprouts have a shorter shelf life than clover leaves because they contain more water and spoil quickly.

Food Transportation

Semanggi snacks that have been produced are transported to the place or location of sale. The results

showed that the containers used to transport the components of *Semanggi* snacks to places of sale or places to serve *Semanggi* snacks were all (100%) clean, strong, intact, of adequate size, and covered with plastic. In contrast, other studies show that all *Pecel* traders transport food not in closed conditions³³.

Food Serving

There were 12 respondents (37.5%) who were selling near the source of the pollutant (dust) because it was next to a busy highway where vehicles were passing by. Nine respondents (28.1%) sold their clover near a garbage pile with many insects (flies). Places to sell near garbage dumps can become nests for insects and nuisance animals because of the dirty location³⁴.

A total of 47 questions measured respondents' sanitation conditions and categorized them into sound, sufficient, and insufficient. Most of the respondents (87.5%) fall into the excellent category (score 32-47) in implementing food processing sanitation (Table 4). Most of the food processing sanitation requirements have met the requirements. This finding follows other studies showing that food sanitation hygiene is mainly in the excellent category³⁴. However, there are still some aspects that are lacking, such as material storage areas that have not been arranged in shelves according to type, the roof is not watertight and leaking, the ceiling is not brightly colored, there is a ceiling with holes, there is no place for washing hands, there is no closed trash in the kitchen.

Table 3. Respondents' sanitary conditions related to the presence of *Escherichia coli* bacteria in clover

Rating items	Yes		No		Total	
	n	%	n	%	N	%
Selection of Raw Materials						
Clean Condition						
It is maintained and looks clean, with no dust	25	78.1	7	21.9	32	100
Placement of Raw Materials						
Placement separate from the finished food	31	96.9	1	3.1	32	100
Storage						
Raw materials are stored and arranged in similar/similar shelves	1	3.1	31	96.9	32	100
The raw materials that come in first are used first	30	93.8	2	6.3	32	100
The distance between the food and the floor is 15 cm	24	75	8	25	32	100
The distance between the food and the wall is 5 cm	27	84.4	5	15.6	32	100
The distance between the food and the ceiling is 60 cm	30	93.8	2	6.3	32	100
Food Processing Site						
Condition of floors, walls, roofs, ceilings, and doors						
The floor is non-slip, flat, and waterproof	27	84.4	5	15.6	32	100
The wall surface is even and smooth	23	71.9	9	28.1	32	100
The roof is watertight, does not leak, is quite sloping, and does not become a nest for rats and insects	21	65.6	11	34.4	32	100
The ceiling surface is flat and light in color	12	37.5	20	62.5	32	100
The ceiling is not perforated	11	34.4	21	65.6	32	100
The door looks strong and can be closed tightly.	32	100	0	0	32	100
Easy-to-open door (towards the outside)	5	15.6	27	84.4	32	100
Kitchen Conditions						
Have ventilation or chimney	16	50	16	50	32	100
There is a place to wash hands, a place to wash food, and or a place to wash equipment	0	0	32	100	32	100
There is a trash can made of strong and closed material	2	6.3	30	93.8	32	100
Sound wastewater disposal system (strong and closed material)	22	68.8	10	31.3	32	100
Avoid insects and mice	25	78.1	7	21.9	32	100
Food Processing Equipment						
Looks clean and robust (no damage or cracks)	27	84.4	5	15.6	32	100
Equipment that has been used is washed with clean water and soap	32	100	0	0	32	100
Washed equipment is dried with a tumble dryer or clean cloth	0	0	32	100	32	100
Do not reuse equipment designed for single use only	3	9.4	29	90.6	32	100

Rating items	Yes		No		Total	
	n	%	n	%	N	%
The equipment is made of food-grade material (the surface is not easy to peel off and does not rust)	28	87.5	4	12.5	32	100
Water						
The source of clean water used is at least 10 meters from the pollutant source (Septic tanks, latrines, cattle pens)	31	96.9	1	3.1	32	100
If the water source comes from a well, the construction of the well must meet the requirements.						
The height of the lip of the well ≥ 80 cm	31	96.9	1	3.1	32	100
Well walls are watertight up to ≥ 3 meters	32	100	0	0	32	100
The location of the cloverleaf is dried in the sun						
Dust free	4	12.5	28	87.5	32	100
Free from insects (flies) or other animals	26	81.3	6	18.8	32	100
Food storage						
Closed, protected from dust, harmful chemicals, insects, rats and/or other animals	3	9.4	29	90.6	32	100
Storage containers look clean and safe, and lids look clean	32	96.9	0	0	32	100
Separate storage	30	93.8	2	6.3	32	100
Presentation						
The container used for serving (banana leaves) is in good condition, clean	32	100	0	0	32	100
The equipment used is clean	32	100	0	0	32	100
Selling location:						
Located at least 500 meters from pollutant sources such as dust, smoke, TPA/TPS, public toilets	20	62.5	12	37.5	32	100
Babab from insects (flies)	23	71.9	9	28.1	32	100

Table 4. Respondent's personal hygiene and sanitation categories related to the presence of Escherichia coli bacteria in clover

Category	Amount (n)	Percentage (%)
Personal Hygiene		
Good	19	59.4
Enough	13	40.6
Not enough	0	0
Total	32	100
Sanitation		
Good	28	87.5
Enough	4	12.5
Not enough	0	0
Total	32	100

Presence of E. Coli Bacteria in Semanggi Snacks

Escherichia coli bacteria are often used as a marker for the presence of microorganisms in food. According to the Decree of the Minister of Health of the Republic of Indonesia No. 1096 of 2011¹² concerning Sanitation and Hygiene Foodservice, food consumption must be protected from bacterial contamination such as E. Coli. The germ count for bacteria must be 0 (zero), or there may not be any E. Coli bacteria in the food consumed. Laboratory test results showed that out of 64 samples consisting of 32 clover leaf samples and 32 seasoning samples, there were 2 (two) positive samples of Escherichia coli bacteria consisting of one clover leaf sample (respondent no. 24) and one spice sample (respondent no. 22).

Good knowledge related to food hygiene and sanitation is expected to make customers carry out food sanitation hygiene properly. With the implementation of good food sanitation hygiene, it is expected to produce quality food safe from contamination. The results showed that most of the knowledge level of the handlers was in the high category, namely 96.9%. However, E. Coli bacteria were still found in the cloverleaf and seasoning samples. During the research, it was seen that several respondents had not implemented good and correct sanitation hygiene when processing cloverleaf snacks. This can happen possibly due to behavior that has become a habit. If it is not balanced with awareness, good knowledge will result in poor hygiene implementation³⁵.

In the pilgrim respondents who obtained the seasoning or cloverleaf positive for containing E. Coli

bacteria, it turned out that the total score for applying personal hygiene was in a suitable category. Other research shows a significant relationship between the personal hygiene behavior of food handlers and the content of E. Coli in food. The personal hygiene behavior of handlers who do not meet the requirements has a risk of 8.5 times higher for containing E. Coli in food compared to the behavior of food handlers who meet the requirements³⁶. When the research was conducted, it was found that the condition of the nails of 22 respondents was dirty and long. Long and dirty nails have the potential to cause food contamination. This condition can happen primarily while pounding sweet potatoes to be used as a condiment for clover snacks. In addition, Respondent 22 and Respondent 24 did not correctly apply personal hygiene, properly not washing hands before and after serving clover snacks, not wearing aprons and head coverings, and scratching limbs when preparing clover snacks. This can also have an impact on the occurrence of contamination of food.

Based on the research, one sample of spices that were positive for E. Coli bacteria was found in 22 respondents who applied adequate food processing sanitation. Meanwhile, one sample of cloverleaf that was positive for E. Coli bacteria was found in 24 respondents with good food processing sanitation. In the case of *Semanggi* snacks, the spices and cloverleaf leaves were vivacious for E. Coli bacteria, both of which had food storage areas that were not adequately covered, namely using only banana leaves, plastic, and or cloth napkins. So that when it is not entirely closed, clover leaves and spices can potentially be contaminated with bacteria through the gap in the lid. The location where the two respondents were selling was where there were flies because, around the place where they were selling, there were piles of uncovered trash³⁷.

As for the 24 respondents who carried out good processing sanitation, but E. Coli was still found on cloverleaf leaves, it is possible that it came from outside the factor of implementing food processing sanitation; namely, handlers had not implemented proper personal hygiene, such as not washing their hands with soap before and after serving customers, not wearing aprons or work clothes when selling, and the habit of scratching limbs when serving food. In addition, when observing, the respondents were seen cleaning banana leaves using a cloth or napkin before being used as a container for serving cloverleaf snacks to buyers. The cloth used may be contaminated with bacteria because it has been used repeatedly, and the cloth used to clean banana leaves looks shabby. In line with other research studies, it was stated that E. Coli bacteria were found in the respondent's herbs that met the requirements for food storage. This is because the cloth has been contaminated with bacteria. The cloth has been used repeatedly to clean bottles of herbal medicine and to clean hands when making herbal medicine³⁸.

Based on the results of the interviews, the 22 respondents whose seasonings were positive contained E. Coli bacteria using boiled water or water that they cooked themselves to puree the seasoning for clover snacks. The use of this water also makes the seasoning potentially contaminated with E. Coli. In line with other

studies, it shows that even though traders have good hygiene practices, drinks are still contaminated with *Escherichia coli* bacteria³⁹. This can happen because it is not sufficiently cooked during the water treatment process until it boils so that the bacteria in the water do not die. The raw water used by PDAM Surabaya is river water from Kali Surabaya.

Meanwhile, the condition of the Surabaya River contains many organic and industrial pollutants⁴⁰. According to the World Health Organization (2005), microorganisms can survive in food if the heating and cooking processes are inadequate⁴¹. In line with other studies, it states that if water does not go through a cooking process or is cooked at a temperature that allows E. Coli bacteria to survive⁴².

CONCLUSIONS

Most of the handlers are aged 40 - < 60 (middle age) and are female. The last education is elementary school graduation/equivalent, and they have been trading for over five years, and most traders trade at the exact location. Most respondents have a high level of knowledge about food sanitation and hygiene. The way of processing is done traditionally. Personal hygiene is mainly in the good category, and food processing sanitation is in the excellent category. There were two positive samples of E. Coli bacteria. One positive sample for E. Coli bacteria was found in handlers with high knowledge and exemplary food processing sanitation implementation.

ACKNOWLEDGEMENTS

The researcher would like to thank all the respondents, Faculty of Health Universitas Jember and LP2M of Universitas Jember, for the study support.

Conflict of Interest and Funding Disclosure

All authors have no conflict of interest in this article. LP2M University of Jember funded this research.

REFERENCES

1. Armaid, D. Epidemiologi Penyakit Menular dan Penyakit Tidak Menular. *Jambi Med. J.* **4**, 195–202 (2016).
2. Muna, F. & Khariri. Bakteri Patogen Penyebab Foodborne Diseases. *J. Uin Alauddin* **6**, 74–79 (2020).
3. Mundiatur & Daryanto. Pengelolaan Kesehatan Lingkungan. in *Pengelolaan Kesehatan Lingkungan* (Gaya Medika, 2020).
4. Ministry of Health of Indonesia. *Hygiene Sanitasi Makanan dan Minuman*. (2004).
5. Moelyaningrum AD. Boric Acid and Hazard Analysis Critical Control Point (HACCP) on Kerupuk to Improve the Indonesian's Traditional Foods Safety. *Int. J. Sci. Technol. Res.* **8**, 50–54 (2019).
6. Moelyaningrum AD. Hazard Analysis Critical Control Point pada Produk Tape Singkong untuk Meningkatkan Keamanan Pangan Tradisional Indonesia (Studi di Wilayah Kabupaten Jember). *Indonesia. J. Heal. Sci.* **3**, 41–49 (2012).
7. Ristianingrum, C. T., Moelyaningrum, A. D. &

- Pujiati, R. S. *Hygiene Sanitation and Rhodamin B Dyes in Cenil. J. Heal. Sci. Prev.* **2**, 67–77 (2018).
8. Imansari DS, AD Moelyaningrum & PT Ningrum. *Hygiene Sanitasi dan Kandungan Pewarna Berbahaya pada Keripik Pisang (Studi pada Industri Rumah Tangga Keripik PPisang Kecamatan X Kabupaten Y).* *Amerta Nutr.* **2**, 1–9 (2018).
9. Dayanti S.B, A.D Moelyaningrum. & Ellyke. *Hygiene Sanitasi dan Kandungan Formalin pada Usus Ayam di Pasar Tradisional Kabupaten Jember.* *J. Public Heal. Recode* **4**, 61–70 (2020).
10. Handayani, N. M. A., Adhi, K. T. & Duarsa, D. P. Faktor yang Mempengaruhi Perilaku Penjamah Makanan dalam Penerapan Cara Pengolahan Pangan yang Baik pada Industri Rumah Tangga Pangan di Kabupaten Karangasem Factors Associated with the Behavior of Food Handlers in the Application of Good Manufacturing Pr. *Public Heal. Prev. Med. Arch.* **3**, 194–202 (2015).
11. Yunus, salma P., Umboh, J. M. . & Pinontoan, O. Hubungan Personal *Hygiene* dan Fasilitas Sanitasi dengan Kontaminasi *Escherichia Coli* pada Makanan di Rumah Makan Padang Kota Manado dan Kota Bitung. *J. Ilmu Kesehat. Masy. Unsrat* **5**, 210–220 (2015).
12. Ministry of Health of Indonesia. *Permenkes RI No 1096/MENKES/PER/VI/2011 Tentang Hygiene Sanitasi Jasaboga.* (2011).
13. Marlinda M, AD Moelyaningrum & Ellyke. Keberadaan Bakteri *Eschericia Coli* dan *Coliform* pada Sumur Gali dan Bor Rumah Pemotongan Hewan (RPH). *J. Kesehat. Lingkung. dan Apl. Tek. kesehtan Lingkung.* **16**, 679–688 (2019).
14. Badan Pemeriksaan Obat dan Makanan dan. *Laporan Tahunan 2019. BPOM RI* (2020).
15. Health Office of Surabaya. *Profil Kesehatan Kota Surabaya Tahun 2018.* (Dinas Kesehatan Kota Surabaya, 2019).
16. Health Office of Surabaya. *Profil Kesehatan Kota Surabaya Tahun 2019.* in (Dinas Kesehatan Kota Surabaya, 2020).
17. Megatsari, H., Laksono, A. D. & Moelyaningrum, A. D. Diarrhea Prevalence in East Java, Indonesia: Does Access to Sanitation and Health Behavior Ecologically Related? *Indian J. Forensic Med. Toxicol.* **15**, 1276–1283 (2021).
18. Health Office of Surabaya. *Profil kesehatan Dinkes Kota Surabaya Tahun 2016.* (2017).
19. Dharmawati, I. G. A. A. & Wirata, I. N. Hubungan Tingkat Pendidikan, Umur, dan Masa Kerja dengan Tingkat Pengetahuan Kesehatan Gigi dan Mulut pada Guru Penjaskes SD di Kecamatan Tampak Siring Gianyar. *J. Kesehat. Gigi* **4**, 1–5 (2016).
20. Fatmawati, S., Rosidi, A. & Handarsari, E. Perilaku *Hygiene* Pengolah Makanan Berdasarkan Olahraga Pelajar Jawa Tengah. *Pangan dan Gizi* **04**, 45–52 (2013).
21. Ministry of Health of Indonesia. *Keputusan Menteri Kesehatan Republik Indonesia Nomor 942/Menkes/SK/VII/2003.* (Menteri Kesehatan Republik Indonesia, 2003).
22. Triandini, F. A. & Handajani, S. Pengetahuan, Sikap Penjamah Makanan dan Kondisi *Hygiene* Sanitasi Produksi Otak-Otak Bandeng di Kabupaten Gresik. *E-Journal Boga* **04**, 27–36 (2015).
23. Prianto, M. D. Hubungan *Hygiene* Penjamah Makanan dengan Kualitas Biologi Brem pada Home Industry Brem Desa Kaliabu. *J. Ilm. Keperawatan* **3**, 50–61 (2017).
24. Desiyanto, F. A. & Djannah, S. N. Efektivitas Mencuci Tangan Menggunakan Cairan Pembersih Tangan Antiseptik (Hand Sanitizer) terhadap Jumlah Angka Kuman. *J. Kesehat. Masy. (Journal Public Heal.* **7**, 75–82 (2013).
25. Kasim, K. P. & Sari, A. A. M. S. Hubungan Personal *Hygiene* Penjamah Makanan dengan Kualitas Bakteriologis MPN Coliform pada Jajanan di Wilayah Pasar Segar Panakukang Kota Makassar. *Sulolipu Media Komunikas Sivitas Akad. dan Masy.* **18**, 530–139 (2018).
26. Anggraeni, I. A. & Moelyaningrum, A. D. *Hygiene* Sanitasi dan Kandungan Boraks pada Puduk (Studi di Industri X Kecamatan Gresik Kabupaten Gresik). *Bul. Keslingmas* **41**, 11–22 (2022).
27. Ministry of Health of Indonesia. *Keputusan Menteri Kesehatan Republik Indonesia Nomor 715/Menkes/SK/V/2003. Tentang Persyaratan Hygiene Sanitasi Jasaboga.* (Menteri Kesehatan Republik Indonesia, 2003).
28. Sonia, V., Koesyanto, H. & W, A. S. Evaluasi Penerapan *Hygiene* dan Sanitasi Penyelenggaraan Makanan di RSUD Sunan Kalijaga Kabupaten Demak Tahun 2013. *Unnes J. Public Heal.* **4**, 124–131 (2015).
29. Mashudi, M. Analisis *Hygiene* Dan Sanitasi Kantin FPSB Universitas Islam Indonesia. (Universitas Islam Indonesia, 2018).
30. Agustiningrum, Y. Hubungan *Hygiene* Sanitasi dengan Angka Kuman Peralatan Makan pada Pedagang Makanan Kaki Lima di Alun-Alun Kota Madiun. *Skripsi (STIKES Bhakti Husada Mulia Madiun., 2018).*
31. Moelyaningrum, A. D., Ningrum, P. T. & Utomo, B. T. E. *Coli* pada Sumber Air DAN Kondisi Sanitasi Terminal Tawang Alun Kabupaten Jember. *IKESMA* **10**, 161–172 (2014).
32. Muhammad, Dewi, E. N. & Kurniasih, R. A. Oksidasi Lemak pada Ikan Ekor Kuning (Caesio cuning) dengan Konsentrasi Garam yang Berbeda. *J. Ilmu dan Teknol. Perikan.* **1**, 67–75 (2019).
33. Sembiring, G. G., Dharma, S. & Marsaulina, I. Penilaian *Hygiene* dan Sanitasi Penjualan Makanan Pecel dan Pemeriksaan Bakteri *Salmonella* di Kecamatan Medan Helvetia 2015. *Dep. Kesehat. Lingkung.* **1**, 1–9 (2015).
34. Trigunarso, S. I. *Hygiene* Sanitasi dan Perilaku Penjamah Makanan dengan Angka Kuman pada Makanan Jajanan di Lingkungan Sekolah. *J. Kesehat.* **11**, 115–124 (2020).
35. Sofiana E. Hubungan *Hygiene* dan Sanitasi dengan Kontaminasi *Escherichia Coli* pada Jajanan di Sekolah Dasar Kecamatan Tapos Depok.

- Kesehatan Masyarakat* (2012).
36. Mauliku, N. E., Budiana, T. A. & Yulianti, R. Hubungan Perilaku Penjamah Makanan dengan Kandungan Bakteri *Escherichia coli* pada Makanan di Unit Dietary Food Service. *PINLITAMAS 1* **1**, 411–415 (2018).
 37. Islamy, G. P., Sumarmi, S. & Farapti, F. Analisis *Hygiene* Sanitasi dan Keamanan Makanan Jajanan di Pasar Besar Kota Malang. *Amerta Nutr.* **2**, 29–36 (2018).
 38. Sarah, Y. Analisis Personal *Hygiene*, *Hygiene* Sanitasi Pengolahan dan Pemeriksaan Kandungan *Escherichia coli* pada Jamu Gendong di Kecamatan Padangsimpuan Selatan Tahun 2019. (Sumatera Utara, 2019).
 39. Afriyanti, L. N. Faktor yang Berhubungan dengan Keberadaan *Escherichia coli* pada Minuman di Kantin Sekolah Dasar Kota Semarang Tahun 2018. (Universitas Negeri Semarang, 2019).
 40. Yudo, S. & Said, N. I. Kondisi Kualitas Air Sungai Surabaya Studi Kasus: Peningkatan Kualitas Air Baku PDAM Surabaya. *J. Teknol. Lingkung.* **20**, 19–28 (2019).
 41. World Health Organization. *Sanitation and Hygiene Promotion*. http://www.who.int/water_sanitation_health/hygiene/sanhygpromo.pdf. (2005).
 42. Julia, P., Latumeten, N. C. & Souisa, G. V. Analisis Cemarkan *Escherichia Coli* pada Jajanan Gorengan dan Minuman Olahan di Depan Kampus Universitas Kristen Indonesia Maluku (Ukim) Ambon. *2-Trik Tunas-Tunas Ris. Kesehat.* **7**, 149–156 (2017).