

**IDENTIFICATION OF ESCHERICHIA COLI BACTERIA ON FRUIT ICE
SOLD BY STANDARD TRADERS IN GORONTALO CITY**

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ABSTRACT

Escherichia coli is a normal bacterial flora that is often found in the human intestine, is unique because it can cause primary infections such as diarrhea.

This study aims to identify or see the presence or absence of *Escherichia coli* bacteria in fruit ice sold by street vendors in Gorontalo City.

Methods in research it uses a qualitative approach. The type of research used is descriptive observational where research on the presence of *Escherichia coli* bacteria is carried out on fruit ice drinks according to the results of laboratory tests carried out. The sampling technique used is random sampling and there are 8 locations of fruit ice sellers in Gorontalo City.

Research result What has been done is that all samples that have been examined are not contaminated with *Escherichia Coli* bacteria but are contaminated or contaminated by other pathogenic bacteria.

Keywords: Bacteria, fruit ice, *Escherichia coli*.

INTRODUCTION

Indonesia is a country that has a variety of cultures and ethnic groups, each region has its own characteristics of customs, language, and food. The wealth of Indonesia is quite extensive, the resulting diversity varies which makes our Indonesia richer. It is not enough to go through natural resources alone to include the culinary diversity found in each region that forms the criteria for each region. This culinary diversity can be seen through the most types of food available [21].

Food is needed by residents in the survival of life. The issue of food is one of the most important basic needs, in addition to housing, clothing, education and health. Consuming foods that contain more energy and nutritional compounds that the body needs can affect the nutritional status of individuals. For adolescents, efforts are always carried out

in improving health with nutritional status [3].

The factors that determine the quality of good food can be seen through various aspects, where aspects of delicacy, the content of nutritious compounds in food, and aspects of public health. Food that is interesting, delicious, also has a lot of nutrition, may not mean anything at all if it is not safe to consume it. In this regard, it can be caused because food can be intermediary or substrate in the life of pathogenic microorganisms with other organisms due to abnormalities [3].

Nowadays, street food is widespread in the community, especially for street vendors. According to the Foods and Agriculture Organizations (FAO), street food is defined as food and beverages prepared and sold through street vendors on the streets and in other public crowded areas that are directly

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consumed or eaten, neither processed nor prepared, which is highly sustainable [5]. . Food can trigger disturbances, certain things are caused by food which has the potential to cause disturbances to the community if in its manufacture, certain foods have not been processed in an appropriate and correct way [8].

One type of food disorder is diarrhea. Diarrheal disorders remain a public health problem in developing countries, including Indonesia, because they always appear in the form of Extraordinary Events (KLB) and coincide with large deaths [1].

Based on the World Health Organization (WHO), diarrheal disorders are still a global problem in terms of high morbidity and mortality in several countries, especially in Indonesia. It is stated that more than 2 billion cases of diarrhea occur in the world every year, until deep, diarrhea is included in the 10 largest deaths, is in the 9th level of the death rate of 1.5 million cases of all age groups. Through this information, it was also found that the countries with the most frequent cases of diarrhea were: Bangladesh (72.4% of cases), Kiribati (61.3% of cases), Sierra Leone (85.3% of cases), and Zimbabwe (35.8% of cases). case).

Based on WHO data, namely, information was also obtained which revealed that the average number of diarrheal disorders in children under 5 years old was 499,000 people. WHO information reveals, reaching 1.7 billion cases of diarrhea in children with a mortality rate of 760,000 for children under five every year WHO, but for adults and the elderly (over 60 years), the prevalence is 557,000 cases at a mortality rate of 228,047 each year. [18]. In Indonesia, based on Risesdas, the prevalence of diarrhea is 1,017,290 cases. The province with the highest prevalence of diarrhea was West Java Province in the

number of 186,809 cases, and the province with the lowest prevalence was North Kalimantan Province in most cases totaling 2,733 cases. And Gorontalo Province is ranked 32 in most cases totaling 4,574 cases. Based on the Risesdas data, information was also obtained where the age group that most frequently experienced diarrhea was the age group 5 to 14 years, amounting to 182,338 cases, but the age group with the lowest incidence of diarrhea was the age group under 1 year which amounted to 18,225 cases.

Based on data from Gorontalo Province, Indonesian health, the prevalence of diarrhea in Gorontalo Province is 16,667 cases. Through these data, information is generated where the highest cases are in the District (Kab.) Boalemo which has around 4,390 cases, followed by Gorontalo City with 3,641 cases, followed by Kab. Bone Bolango totaled 3,229 cases, Kab. Pohuwato totaled 2,633 cases, Kab. Gorontalo totaled 1,532 cases and the last one was Kab. North Gorontalo totaled 1,239 cases. The formation of foodborne disorders, such as diarrhea, is very high in Indonesia due to biological pollution, which is 36.7%. A biological contamination that can expose food is the presence of coliform bacteria. Bacteria of the Enterobacter, Shigella, Proteus, Salmonella and Escherichia coli groups include a variety of Coliform bacteria [20].

The presence of Escherichia coli bacteria with water or food sources can be an indication of the formation of contaminated feces. The presence of Escherichia coli in water and food is also known to have the largest correlation in the discovery of pathogenic seeds in food. Escherichia coli or often referred to as E. coli, is a kind of gram negative bacterial species. This can be caused by various circumstances, the main impact being in

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the manufacture of certain foods and beverages, for example: processing stages, personal hygiene for traders, equipment sanitation hygiene, raw product sanitation hygiene, processing stages and sales facilities [1].

One of the foods/drinks that quickly spread the presence of Escherichia coli is fruit iced drinks. In addition to certain effects, fruit ice is more susceptible to Escherichia coli bacterial contamination, this is due to the presence of fruit with the lowest acid content, with the possibility of pathogenic bacteria surviving. In addition, the temperature through fruit ice contained in the optimum temperature for bacterial life is 10°C. the optimum temperature for the development of Escherichia coli bacteria is in the range of 10-60°C [10].

In addition, fruit ice includes the fresh food group whose manufacture has not passed the cooking stage first. Until, if it is not made properly by standardization of hygiene and sanitation, it can potentially contaminate the Escherichia coli bacteria. According to observations made by Sofiana, it was identified that 51.4% of snack drinks were contaminated with various Escherichiae coli bacteria. Likewise, the observation data carried out by Nadanti in 29 samples of fruit ice, where the results obtained were 26 (89.6%) samples of fruit ice contained Coliforme bacteria by passing the threshold.

Because fruit ice snacks have the potential for microbial contamination, observers feel it is necessary to conduct a study on the identification or presence of Escherichia coli bacteria against fruit ice traders in Gorontalo City.

Pre Analytical :

Lactose Broth (LB) [24].

- a) Weigh the liquid media as much as 0.65 grams in a 100 mL Erlenmeyer container that has been labeled using a balance.

- b) Dissolving using distilled water in the amount of \pm 50 mL.
- c) Check the pH, when it is very basic, add HCl and when it is very acidic, add NaOH solution until it is based on the expected pH.
- d) Pipetting of liquid media in a screwed tube with a complete label of 5 mL (not aseptic).
- e) Durable screw tube is stored in a glass cup then covered with newspaper and then tied with rope.
- f) Put it in an autoclave using a pressure of 15 Psi (Pound Square Inch), a temperature of 121°C for up to 15 minutes
- g) *Eosin Methylene Blue* (EMB) [23].
 - a. Weighing 37.5 grams (or adjustments in EMB Media bottles) of EMB Media powder, liquid using 1 liter of aquadest.
 - b. Heating until boiling in dissolving media.
 - c. Sterilized in an autoclave at a temperature of 121°C for up to 15 minutes.
 - d. Wait until the temperature is warm (45°C-50°C), homogenized.
 - e. Pour into sterile petri dishes.

Escherichia coli and Bacterium coli commune are bacterial names taken by naming the person who got it, Theodore Escheriche. During this period, he gave the name Escherichia coli to Bacterium coli mutabilae [19].

Escherichia coli is a normal bacterial flora that is always found in the human intestine, it is unique and can cause primary infections such as diarrhea [12].

Based on Pelczar (2013), the classification of Escherichia coli include:

Kingdoms: *bacteria*
phylum: *Proteobacteriae*
Class: *Gamma Proteobacteriae*
Order: *Enterobacterialesh*
Family: *Enterobacteriaceae*
Genus: *Escherichia*

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Species: *Escherichia coli*.

Escherichia coli (*E. coli*) is a gram-negative bacterium that forms short rods or is always called coccobacillus. In gram-negative bacteria, these are bacteria that have not maintained the crystal violet color compound when Gram staining is carried out so that it can turn red when viewed under a microscope [17].

Escherichia coli has flagella, has a size of 0.4-0.7 μ m x 1.4 μ m and has storage [14]. *E. coli* has a length of about 2 μ m, a diameter of 0.7 μ m, a width of 0.4-0.7 μ m, which is facultatively anaerobic. With colonial circular shape, convex, and smooth the edges are markedly [10].

Escherichia coli including bacteria that have 150 antigenic O types, 50 antigenic H types, as well as 90 antigenic K types. Various O antigenic can be carried in the presence of other microorganisms, to resemble those that have Shigellae. Sometimes the disturbance is specifically related to O antigen, it can be seen in urinary tract infections as well as diarrhea [12].

Escherichia coli including facultative anaerobic bacteria that can grow under aerobic or anaerobic conditions. Oxygen is used in the carbon source through the outside which is useful for energy in the best life in an oxidative way. Anaerobic life uses fermentation to obtain energy for its survival [19].

Escherichia coli form pathogenic when the amount exceeds optimally present in the body. These bacteria also acquire enterotoxins to cause diarrhea [11]. The number of *Escherichia coli* strains to be evolved and then obtained the virulence power to be able to infect the host. In a wide variety of pathogenic *Escherichia coli* can cause infection in the urinary tract. There are various types of pathogens including:

ETEC is a pathogenic *Escherichia coli* caused mainly by acute diarrhea in dehydration in children and adults in

countries with 2 or 3 seasons. ETEC obtains enterotoxins to cause the formation of excretion of the body's electrolyte solution until diarrhea occurs in dehydration. In an immunological manner the enterotoxins obtained by ETEC are appropriate in the enterotoxins to be obtained by *V. cholerae*. Enterotoxin ETEC is composed of two types, namely:

Labile Toxine (LT) which has the largest molecular weight and is not yet immune to heating (killed at 60°C for up to 10 minutes); The toxin corresponds to the cholerae toxin.

Stabile Toxine (ST) is a small peptide composed of 18-48 amino acids with a large number of cysteine chains. It has the lowest molecular weight, is immune to heating and does not yet have antigenic properties. Humans can play a role as carriers of these germs, where to carry germs but he himself is not sick.

Transmission of germs can be done by means of food-borne or waterborne. In areas where diarrhea is endemic, such as Indonesia, ETEC is also a cause, especially acute diarrhea, according to cholerae as well as traveller's diarrhea [11].

EPEC (Enteropathogenic *Escherichia coli*), including the initial strain of *Escherichia coli* strains, has been successfully identified for causing pathogenic diarrhea in infants and children in hospitals in the UK and various countries in Europe. In various urban areas, about 30% of acute diarrhea problems in infants and children are caused by EPEC. The mechanism for the formation of diarrhea caused by the presence of EPEC cannot be clearly stated, but it is suspected that the EPEC obtained a cytotoxin which is included as a result of the formation of diarrhea. Diarrhea disorders that appear are generally self-limited but can be fatal or develop into persistent diarrhea, especially for children under 6 months of age. In

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developing countries, children with EPEC infection are generally one year old or older [7].

EIEC has various acidifications against Shigellae including on the matter of biochemical reagents in short sugars, serological which are pathogenic. When it comes to Shigellae, EIEC applies intestinal mucosal penetration which performs its multiplication in the epithelial cells of the colone (large intestine). The breakdown by forming in the intestinal epithelium results in bloody diarrhea. In a microscopic manner, polymorphonuclear leukocytes were still present in the faeces of patients with infected EIEC. Clinical symptoms that appear such as dysentery to be caused by the presence of Shigellae [9].

The pathogenicity of EHEC is in producing cytotoxins by being responsible for the formation of inflammation and bleeding to spread to the large intestine in causing the formation of haemolytics ureamics syndromes, especially in children. The criteria for symptoms to appear with signs of acute diarrhea, convulsions, fever and relatively fast duration of diarrhea become bloody. The occurrence of bloody diarrhea was specific for comparing EHEC and Shigellae strains. In developing countries the occurrence of diarrhea due to EHEC remains rare.

EAEC has been obtained in various countries in the region. Transmission can be food-borne or water-borne. The pathogenicity of EAEC is formed because the bacteria are tightly attached to the intestinal mucosa to cause abnormalities. The mechanism by which diarrhea develops as a result of EAEC is not well known, but predicts the acquisition of cytotoxins to cause diarrhea. Various EAEC strains have serotypes such as EPEC. EAEC causes watery diarrhea in children and can progress to persistent diarrhea [7].

Escherichia coli can live excessively when someone uses food that has been contaminated by certain bacteria such as milk, food that has not been processed perfectly, or food and drink contaminated with feces [11]

These bacteria can be pathogenic when present in large quantities in the human body. *Escherichia coli* can live in high temperatures as well as the lowest, in the lowest temperature of 7 ° C in the highest temperature to 44 ° C. but the bacteria *Escherichia coli* live normally at a temperature of 35-37°C in a pH of 7-7.5. It grows in humid areas and can die when the process of heating food is formed [22]. Sources of bacterial contamination of *Escherichia coli* can come from several vehicles and places, the following include sources of bacterial contamination of *Escherichia coli* which are: 1) contaminated food, beef, especially for immature, 2) unpasteurized milk, 3) fruits and vegetables raw, 4) contaminated water, including drinking water that has not been boiled, 5) feces of infected people [20].

Escherichia coli can live excessively when someone uses food that has been contaminated by certain bacteria such as milk, food that has not been processed perfectly, or food and drink contaminated with feces [7].

These bacteria can also enter the human body from hands or equipment as well as bottles, pacifiers, thermometers, and eating utensils that are contaminated with feces. Exposure can be formed when the worker contacts to make food so that it causes the transmission of food-borne disorders and foodborne diseases. It can also be infected when the individual picks the anal area and then has not washed his hands which then holds everything around him. Also objects and food that have been exposed are taken by other people [22]. Effects to affect bacterial life are: pH, temperature, nutrition, osmotic pressure

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and others. Bacteria have a temperature suitability in their own pH in normal life [12].

a. Temperature

Microorganisms are divided into 3 according to temperature, namely: thermophilic, mesophilic and psychrophilic. Each of them has a different temperature interval [9]. In the next merge:

Table 1 Classification of Microorganisms by Temperature

No	Sifat Mikroorganisme	Suhu Minuman	Suhu Optimum	Suhu Maksimum
1.	Termofilik	40-45°C	55-75°C	60-85°C
2.	Mesofilik	10-15°C	30-45°C	35-47°C
3.	Psikrofilik :			
	a. Fakultatif	5°C	25-30°C	30-35°C
	b. Mutlak	5°C	15-18°C	-22°C

(Source: Harti, 2015)

b. pH

The pH of microorganisms in their development is not the same. In acidophiles (2.0-5.0), neutrophils (5.5-8.0), and alkalophiles (8.4-10.0). Mainly bacteria enter the pH 7-7.5.

c. Nutrition

Nutrients are organic products that break down in the presence of microorganisms that make food for life. The nutrients for use by microorganisms include: Carbone (C), Nitrogen (N), Oxygen (O), minerals and vitamins.

d. Osmotic Pressure

Osmotic pressure can affect the exchange of water in cells. Divided into 3 conditions which are hypotonic, isotonic, and hypertonic [9].

Growth Media Escherichia coli bacteria can live in Endoe agar, Mac Conkays agar, and Eosine Methylent Blhue (EMB) media, these bacteria have strains with microaerophilic properties to

require oxygen in life but not with oxygen and various through Escherichia coli can still grow. 7]. Besides that, it also has aerophilic strains to be able to hemolyze, in Blood Agar Plates (BAP) media these bacteria can hemodialysis in hemodialysis (total hemolysis).

In Eosine Methylent Blues (EMB) media, these bacteria can ferment lactose until the color is shiny green. Bacteria by fermenting lactose briefly then obtaining acid also decreases the pH. This causes the colonies on certain media to have a shiny green color, for example Escherichia coli. Bacteria that have not fermented lactose can add to the pH and protein. So that the colonies can not be colored [20].

Hygiene Sanitary food and beverages are food products that are more needed for residents, which are useful in the continuity of their lives. The best food in life or sustain life. Food produces energy with products that are needed in the construction and replacement of damaged tissues, in work and in the maintenance of the body's defenses through disorders [2].

Food can make people healthy or sick. So that it is always handled and managed properly so that it is useful for the body. Appropriate and correct management in general is managing food and beverages in accordance with the rules in food sanitized hygiene procedures [14].

Ice fruit is a drink of the freshest fruit juice, a kind of shopping for basic products through fruit, which is often consumed by the wider community because it contains nutrients and vitamins that are better for health. The freshest fruit is a type of grocery which is produced without cooking/heating management, the freshest fruit is cleaned using clean water (streamed water), cut into pieces, blended (smoothed) with the addition of water and sugar, these stages range from being

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polluted by physical, chemical, and biological means. biologicals both at the stage of selecting raw products to the stage of presenting. That's why always pay attention to the hygienic aspects of sanitation in HACCP (Hazard Analysis of Critical Control Points) [4].

Food and beverage sanitation hygiene is an effort to control the causes of food, people, premises and equipment to cause or may cause health problems or disorders. Sanitary hygiene requirements are technical requirements specified in restaurant and restaurant materials, personnel and equipment which include bacteriological, chemical and physical requirements [13].

The principle of sanitation and hygiene in food and beverage processing, generally, the management of food and beverages has a sanitary hygiene procedure so that in controlling the

influence of places, equipment, people and food, which can or may cause health problems or food poisoning. There are 6 (six) hygiene procedures for food and beverage sanitation, namely: selecting food products, storing food products, managing food, transporting cooked food and storing cooked food.

Fruit ice is included in fresh food but its processing does not go through the cooking process first. Fruit ice is more susceptible to coliform bacterial contamination, this is because there are fruits that have the lowest acid content which has the lowest probability of survival of pathogenic bacteria. Besides that, the temperature through fruit ice is in the optimum temperature for bacterial growth which is around 10°C. The optimum temperature for the development of coliform bacteria is in the range of 10-60°C [2].

RESEARCH METHODS

Approach This study uses qualitative proximity which aims to identify the presence of Escherichia coli in fruit ice drinks sold in Gorontalo City.

The type of observation used in this observation is descriptive observational where the observation of Escherichia coli bacteria is carried out in fruit ice drinks according to the results of laboratory tests carried out. Types and sources of information to be used in these observations are primary data and secondary data. The primary data in this study is information on the bacteriological condition in this case contamination by Escherichia coli bacteria in fruit ice samples produced through various places that sell fruit ice drinks in Gorontalo City, including documentation of laboratory testing. However, secondary data in this study was obtained through literature study by retrieval of information such as data

such as observational journals that were originally relevant in these observations. The results to be obtained are processed in a descriptive way and then presented in tabular form along with a narration. The variable in this observation is a single variable where the identification of Escherichia coli bacteria in fruit ice drinks.

The observation population is all fruit ice sellers in Gorontalo City. Through field observation data carried out in April 2021, it is known that there are many active fruit ice sellers, only 8 traders.

The sample in this observation is fruit ice sold in Gorontalo City, which has 8 traders.

The sample collection technique is using random sampling technique and there are 8 locations of fruit ice sellers in Gorontalo City.

In this observation, the validity of the data used is by laboratory testing

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using a qualitative approach method by looking at the presence or absence of Escherichia coli bacteria in fruit ice. The way to see the presence of Escherichia coli bacteria is by looking at the color change in the sample that has been examined using the complementary test (EMBA), where a positive sample will change color to greenish red or metallic green, whereas if this sample does not change color then this sample is negative for Escherichia. coli or not contaminated with Escherichia coli bacteria.

RESEARCH RESULT

In accordance with the observational data carried out at the Microbiological Laboratory of Bina Mandiri University, Gorontalo, from 8 samples of Escherichia coli examination on fruit ice in Gorontalo City, the following results were obtained:

1. Coliform Test Results on Lactose Broth . Media

Based on the results and research from the estimator test using lactose broth (LB) media to see the presence of Coliform bacteria with a sign that there is a change in color from green to yellow and the formation of gas in the media is presented in the following table 4.1:

Table 1 Estimator Test Results on Lactose Broth (LB) Media

Kode Sampel	Uji Penduga		
	Positif (+)	Negatif (-)	Nilai MPN
Sampel 1	✓		>2.400
Sampel 2	✓		>2.400
Sampel 3	✓		>2.400
Sampel 4	✓		>2.400
Sampel 5	✓		>2.400
Sampel 6	✓		>2.400
Sampel 7	✓		>2.400
Sampel 8	✓		>2.400

(Source: Primary Data 2021)

Based on Table 1 Old Results Table 4.1 the results of the examination

of coliform bacteria on fruit ice samples on lactose broth (LB) media showed that all samples tested were positive. Bacterial growth in lactose broth (LB) media was characterized by gas formation and turbidity.

2. Test Results on EMBA . Media

Based on the results and research of complementary tests using the media by looking at the presence of Coliform bacteria with signs of colonies in the presentation media in table 4.2 below:

Table 2 Complementary Test Results in Eosine Methylene Blue Agar (EMBA) Media

No	Kode Sampel	Ciri Koloni Pada Media EMBA	Keterangan
1.	Sampel 1	Bulat, Merah muda	Negatif <i>E.coli</i>
2.	Sampel 2	Merah muda	Negatif <i>E.coli</i>
3.	Sampel 3	Merah muda	Negatif <i>E.coli</i>
4.	Sampel 4	Bulat, Merah muda Keunguan	Negatif <i>E.coli</i>
5.	Sampel 5	Bulat, Merah muda	Negatif <i>E.coli</i>
6.	Sampel 6	Bulat, Merah muda	Negatif <i>E.coli</i>
7.	Sampel 7	Merah muda	Negatif <i>E.coli</i>
8.	Sampel 8	Merah muda	Negatif <i>E.coli</i>

(Source: Primary Data 2021)

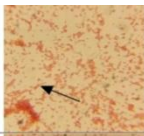
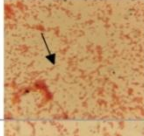
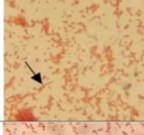
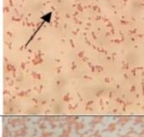
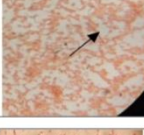
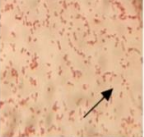
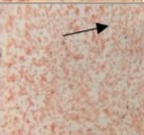
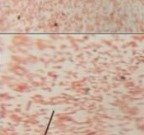
Table 4.2 shows where all the samples to be grown in EMBA media contained bacterial colony growth, namely, round, and purplish pink. However, no metallic green colonies were found, so it can be said that Escherichia coli was negative in the tested samples.

3. Gram Stain Results

In accordance with the observational data that has been carried out, the data from gram staining is presented in the following table:

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Table 3 Results of gram staining of colonies from samples grown on EMBA . media

Sampel 1	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 2	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 3	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 4	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 5	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 6	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 7	Merah	Batang (basil)		Bakteri Gram Negatif
Sampel 8	Merah	Batang (basil)		Bakteri Gram Negatif

(Source: Primary Data 2021)

Table 4.3 gram staining to see the morphology of Escherichia coli bacteria from EMBA media microscopically based on differences in structure whether gram-positive or gram-negative bacteria with the results obtained that the morphology of the bacteria forms a red rod to indicate certain gram-negative bacteria.

DISCUSSION

In accordance with observational data regarding the identification of the presence or absence of Escherichia coli bacteria in fruit ice to exist in Gorontalo City, that fruit ice samples sold in several Gorontalo cities were negative Escherichia coli or not contaminated with Escherichia coli bacteria.

Fruit ice is a type of drink that is made up of pieces of various fruits with sugar, milk, syrup and ice cubes. Processing fruit ice is actually simpler. The delicious taste with fresh fruit ice is the key in the products used. The next thing to choose is that the fruit is always appropriate, of course, the selection of fruit that remains fresh. In fresh fruit so that the vitamin content in certain fruits is maintained in its entirety, so that in consuming fruit ice, health can also be guaranteed [2].

According to Kurniasih et al, contamination of Escherichia coli bacteria in food can be caused by a state of hygiene with sanitation that has not been in the location of food processing. And the opportunity for the formation of certain contaminants is experienced in each stage of food management. Unhygienic food management can cause food products to appear that can cause health problems for buyers and consumers.

Escherichia coli can grow in a variety of locations with conditions including food and drink. The water contaminated with Escherichia coli bacteria cannot be used for food processing needs, such as cooking, washing tableware and washing food products. In this case, the possibility of transferring Escherichia coli through water in food until it can cause irritation [12].

According to Cheptham Media Emba is a selective medium for isolating Escherichia coli bacteria. The EMBA

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contains the carbohydrate lactose, in the presence of this carbohydrate. The color when fertilizing bacteria in EMBA media is purplish red. The color change to metallic green in the media was caused by Escherichia coli bacteria that could ferment lactose to increase the acid content of the media. According to Saidah and Susilawati EMBA media is a selective medium in isolating E.coli bacteria. Certain media can compare Escherichia coli bacteria to other Enterobacteriaceae bacteria. The Eosine Methylene Blue Agar media has a feature where there is lactose content in the media and can be used as a comparison factor for Escherichia coli bacteria against others.

In EMBA media, it can be used to isolate and differentiate enteric and coliform bacteria. EMB media contains lactose so that it can compare bacterial groups in strength when fermenting lactose, bacteria that can ferment lactose include Escherichia coli, these bacteria include bacteria that can ferment lactose briefly and produce a lot of acid so that it can get a purple-black colonial color and a metallic glossy green. .

Gram stain is used to compare bacteria that are Gram positive with Gram negative. Gram staining is carried out through 4 steps, namely applying paint especially (crystal violet), intensifying paint, especially in adding lugole fluid, washing in a laxative liquid (alcohol) by applying a covering paint (safranine). Gram negative. The principle of Gram-negative staining is that bacteria can bind to the last dye carried out in the test. Bacterial Escherichia coli obtained data of red bacterial cells, forming rods. Gram A (crystal violet) staining results in crystal violet which can change the entire surface of the bacterial cell. The addition of Gram B (lugol) resulted in the formation of a violet-lugole crystal

complex which could increase the affinity of color compounds by bacterial cells, the whole bacteria could be purple. Added Gram C (alcohol) results in the formation of pores in Gram negative which have a number of layers of fat (soluble lipids in ethanol), until the crystal violet-Lugol complex has not been attached to the bacterial cell wall, causing Gram negative cells to lack a purple color. Added Gram D (safranine) causes Gram-negative bacterial cells to be absorbed by the dye compound into a red color [16]. Added Gram C (alcohol) results in the formation of pores in Gram negative which have a number of layers of fat (soluble lipids in ethanol), until the crystal violet-Lugol complex has not been attached to the bacterial cell wall, causing Gram negative cells to lack a purple color. Added Gram D (safranine) causes Gram-negative bacterial cells to be absorbed by the dye compound into a red color [16]. Added Gram C (alcohol) results in the formation of pores in Gram negative which have a number of layers of fat (soluble lipids in ethanol), until the crystal violet-Lugol complex has not been attached to the bacterial cell wall, causing Gram negative cells to lack a purple color. Added Gram D (safranine) causes Gram-negative bacterial cells to be absorbed by the dye compound into a red color [16].

Based on the results of the study, it was found that all fruit ice samples in Gorontalo City did not contain or were not contaminated with Escherichia coli bacteria. The factor that causes the fruit ice itself to be free from Escherichia coli is because it goes through a good and correct processing. And there are several other factors that cause fruit ice to be free or not contaminated with Escherichia coli because traders or handlers apply personal hygiene. Compared to previous observations based on observations

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made by Sofiana, it was identified that 51.4% of snack drinks were contaminated with various Escherichiae coli bacteria. Likewise, the research data carried out by Nadanti, in 29 samples of fruit ice, where the results obtained where 26 (89.6%) samples of fruit ice contained Coliforme bacteria by passing the threshold. Furthermore, Siti Mukhlisoh in 2009 in fruit juices sold in the Tembalang sub-district area showed that through 20 samples of fruit juice drinks obtained and tested in the laboratory, they were 100% positive for Escherichia coli bacteria. fruit is not yet the best in the percentage of each 55%, 65%, 30% and 45%. Bacterial content; Escherichia coli in fruit soup, Observational data showed that the bacterial coliforme test of 24 colonies/ml had the threshold determined by SNI, which was 2×10^1 colonies/ml. However, in testing the Escherichia coli bacteria on trader A there was 2.40 / ml, trader B was 11 / ml and for trader C there was 24 / ml. The three samples have passed the threshold determined by the presence of SNI, which is < 3 /ml [22].

Coliforme bacterial contaminants in beverages can cause health problems for consumers [25]. In the average number of Coliforme bacterial contaminants in fruit ice with a range of 8×10^6 colonies/gram, a certain amount exceeds a predetermined threshold of 1×10^4 colonies/gr, certain things can cause health disorders such as infections in the gastrointestinal system such as diarrhea [8].

CONCLUSION

In accordance with the observational data, the Escherichia Coli bacteria identified in fruit ice in the city of Gorontalo that have been carried out can be concluded that all 8 samples were not found or not contaminated with

Escherichia Coli bacteria, only contaminated with other types of pathogenic bacteria.

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