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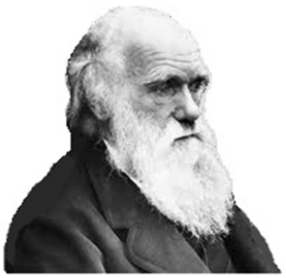

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Isolation and Identification of Potential Pathogenic Bacteria in Living Carp (*Cyprinus carpio* Linnaeus, 1758) Sold in Supermarkets in Cimahi City, Java

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ABSTRACT

This research was conducted with the aim to analyze the presence of potential pathogenic bacteria in carp that are sold live in supermarkets in the city of Cimahi and to find out the species of these bacteria. Fish samples were obtained from two supermarkets in Cimahi City, Transmart and Superindo. From each supermarket three fish samples were taken once a week and repeated three times. Bacteria were isolated from several parts of the fish body namely body surface mucus, gills, liver, and kidneys. The results of isolation from each target organ were biochemically tested to determine the species of bacteria. Potential pathogenic bacteria found in carp from this research are *Aeromonas hydrophila, Aeromonas sobria, Aeromonas schubertii, Aeromonas media, Aeromonas caviae, Aeromonas ecrenophila, Pseudomonas stutzeri, Pseudomonas pseudoalcaligenes, Pseudomonas aeruginosa, Aeromonas caviae, Aeromonas ecrenophila, Pseudomonas stutzeri, Pseudomonas pseudoalcaligenes, Pseudomonas aeruginosa, Plesiomonas shigelloides, Bacillus megaterium, Bacillus thuringiensis, Neisseria mucosa, Citrobacter freundii, Serratia marcescens, Staphylococcus epidermidis, Micrococcus luteus, Enterobacter aerogenes,* and *Enterobacter sakazakii.*

***Keywords*:** Cyprinus carpio, Isolation and identification, Pathogenic bacteria, Supermarket

1. INTRODUCTION

Food fish can be interpreted as all marine and freshwater biodiversity that contain high protein and have important significance for economic interests. Food fish farming activities are carried out to provide the needs of the community for fish consumption. Food fish can be

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marketed after going through the processing stage or sold fresh. It is also not uncommon for food fish to be marketed alive to ensure the freshness of the fish.

Supermarkets are modern markets managed by modern management, generally found in urban areas, as providers of goods and services of good quality and service to consumer [1].

Many people choose to shop for daily necessities at the supermarket because of its relatively cleaner location compared to traditional markets. Food fish is sold in supermarkets with a variety of choices. One type of fish sold in supermarkets is carp (*Cyprinus carpio*).

The purpose of marketing fish in living conditions is to maintain the freshness and quality of the fish, but the fish that are sold live in an aquarium are not always healthy. Transmission of diseases and parasites can occur through several mechanisms, including through direct contact between sick fish and healthy fish, sick fish carcasses or through water [2]. In some supermarkets, live carps are put into an aquarium together with other types of fish, such as tilapia and gouramy.

This can cause injury to the body of the carps due to sharp scratches from other fish and cause disease. Some fish may exhibit symptoms of bacterial infections, such as flaked fins, loose scales, surface swimming, wounds, pale body color and slow movements [3].

Water is a natural habitat for various organisms, including bacteria. Pathogenic bacteria found in water which are also a habitat for fish can threaten the health of fish. Bacteria that attack fish can be pathogenic or cause disease.

Disease-carrying agents from the bacterial group include *Aeromonas* spp., *Streptococcus* spp., *Mycobacterium* spp., *Vibrio* spp., *Enterobacteriaceae*, and *Escherichia coli* O157: H7 [4].

Efforts can be made to determine the types of potential pathogenic bacteria by isolating and identifying the bacteria. Identification is an activity carried out to determine certain types of organisms with stages of observation, testing, recording, and identification based on test results [5]. Isolation and identification of bacteria are carried out with a view to knowing precautions against bacterial diseases so that the quality of fish can be maintained.

2. RESEARCH METHOD

2. 1. Time and Place of Research

This research was carried out in January 2020 to February 2020. This research took place at the Quality Control Fish Quarantine Center (BKIPM) Bandung. The samples to be used were taken from two supermarkets in Cimahi City, namely Transmart and Superindo.

2. 2. Tools and Materials

The tools used are Bunsen burners, inoculating loop, Petri dishes, Dissecting kits, frosted paper, laminar flow cabinets, incubators, refrigerators, slide objects, label paper, ovens, analytical scales, pipettes, autoclaves, gloves, and masks. The materials used are TSA (Tryptone Soya Agar), 18 carp fish bought alive from the supermarkets, Aquadest, Sucrose, Maltose, Lactose, Glucose, Dextrose, Inositol, Sorbitol, Arabinosa, Mannitol, Malonate, Nitrate, Pepton, MR-VP, Gelatin, O / O F (Oxidase / Fermentative), MIO (Motility Indole Ornithine), SCA (Simons Citrate Agar), Urea, LIA (Lysin Iron Agar), TSIA (Triple Sugar Iron Agar), 3% KOH, H_2O_2, Oxidase Paper, Kovack Reagents, MR (Methyl red) reagent, α-naphtol reagent, 40% KOH (VP2 reagent), Nitrate A and B reagents.

2. 3. Tools and Materials Preparation

Research preparations were carried out included sample preparation, instrument sterilization, TSA (Tryptone Soya Agar) making, and fish necropsy. Carp samples used were obtained from two different supermarkets in the city of Cimahi. Three fish were taken from each supermarket with a size of 23-30 cm. The fish are then taken to the Bandung Quality Control Fish Quarantine Laboratory (BKIPM) bacterial laboratory. Repetition is done three times in a period of three weeks. The glasswares are first sterilized by using an oven with a temperature of 165 ºC. The tools are wrapped with paper and then bound using mattress yarn then put into the oven for 10 minutes. Preparation was continued by making TSA media carried out by weighing as much as 40 g TSA media, then the aquadest was measured as much as 1,000 mL, after that TSA media and sterile aquadest was mixed in the Erlenmeyer flask and then homogenized, then put into the autoclave for 20 minutes.

2. 4. Clinical Signs Observation and Necropsy

Clinical symptoms in the body of the fish were observed and recorded. The length and weight of the fish was measured. Dissecting kits were prepared in sterile conditions. Furthermore, fish necropsy was performed. Fish was dissected from anus to operculum using an already sterilized scissor. The dissected fish is then stored into plastic Ziplock and given an A code for samples obtained from Superindo, and B for samples from the Transmart supermarket.

2. 5. Isolation and Inoculation

Tryptone Soya Agar (TSA) is prepared and coded at the bottom of the Petri dish using a pen or marker. Bunsen burners are ignited and the inoculating loop is burned until it turns reddish. The lid of the Petri dish containing the TSA media is opened slightly, then the tip of the burned inoculating loop is affixed to the side of the TSA to cooldown a little bit. The tip of the inoculating loop is then rubbed gently on the surface of the desired body part of the fish. In this research bacteria will be isolated from the liver, gills, and kidneys and then etched on the TSA media. TSA that has been etched then was wrapped using paper and then being put into an incubator at a temperature of 26 ºC and allowed to stand for about 18-24 hours.

2. 6. Bacteria purification

The initial isolation results were removed from the incubator and then stored in Laminar. Differences in the shape and color of the growing colonies were observed. New TSA media were prepared as much in accordance with the types of growing colonies based on differences in color and shape in the initial isolation results. The heated inoculation loop is then poked on a bacterial colony which will be purified from the initial isolation results. Inoculation loop is etched on the new TSA media which will be used for purification. TSA that has been scratched and wrapped using paper, then is being put into an incubator with a temperature of 32 ºC and allowed to stand for about 18-24 hours.

2. 7. KOH String Test, Catalase and Oxidase Test

The incubated TSA is then removed and stored in Laminar. KOH 3%, H_2O_2, and Oxidase strips were prepared. KOH 3% and H_2O_2 are dripped on object slide. Heated inoculation loop

is then etched on the surface of the purified bacterial colony. Bacteria on the loop were then attached to KOH 3%, while circular movements were carried out, then the loop was lifted slowly and observed whether mucus is formed or not. If mucus is formed, bacteria are said to be gram-negative and vice versa, if mucus aren't formed, bacteria are gram-positive.

Furthermore, the bacteria on the loop were then attached to H_2O_2 and observed whether foam is formed or not. If foam forms, the bacteria are said to be catalase positive. Bacteria on the loop are affixed to the *Oxidase strip* and color changes are observed. If the strip changes color to purple, then the bacteria are said to be oxidase positive.

2. 8. Biochemical Test

Tubes containing sugars, malonates, nitrates, MR-VP, peptons, gelatin, O / F, MIO, SCA, LIA, Urea, and TSIA are prepared in a rack as many as the amount of bacteria has been purified. The heated loop is then etched on the surface of the pure bacterial colony. Inoculation loop is dipped in a medium of sugar, malonate, nitrate, MRVP, and peptone while shaking slightly. In the semi-solid media, namely gelatin, O / F, and MIO, the loop is inserted into the middle part of the medium and then removed.

On slanted media, namely SCA and Urea, the loop is scratched with zig zag patterns. In the TSIA and LIA media, the loop is inserted into the center first, then zig zag is scratched. Media that has been planted with bacteria are then put into an incubator and allowed to stand for 18-24 hours.

2. 9. Bacteria identification

After 24 hours, biochemical test results can be read by observing the color changes that occur in each media. The results of further reading are written on the worksheet to be further identified. Identification is done by referring to the books "*Cowen and Steels: Manual for the Identification of Medical Bacteria*" and "*Bergeys's Manual of Determinative Bacteriology*".

3. RESULTS AND DISCUSSION

3. 1. Clinical Signs Observation

In the first and second weeks of sampling, on average the fish showed clinical symptoms in the form of caudal fins and pectoral flakes or torn (**Figure 1a**), hemorrhage in the body and tail (**Figure 1b**), ulcers in the body (**Figure 1c**), as well as loose scales and lesions on the body (**Figure 1d**). Clinical symptoms are shown in fish from both supermarkets where sampling is taken.

Fish samples from both supermarkets were taken in the first and second week, weighed between 234-691 grams. During necropsy, fish taken in the first and second weeks have gonad sizes that meet ½ - 2/3 of the abdominal cavity and eggs which are clearly visible in female fish. In contrast to fish in the first and second week, fish taken in the third week show no clinical symptoms.

Weight of fish ranging between 195 - 313 cm and length 23-25.5 cm was performed. Fish taken in the third week have small size and not yet developed gonads.

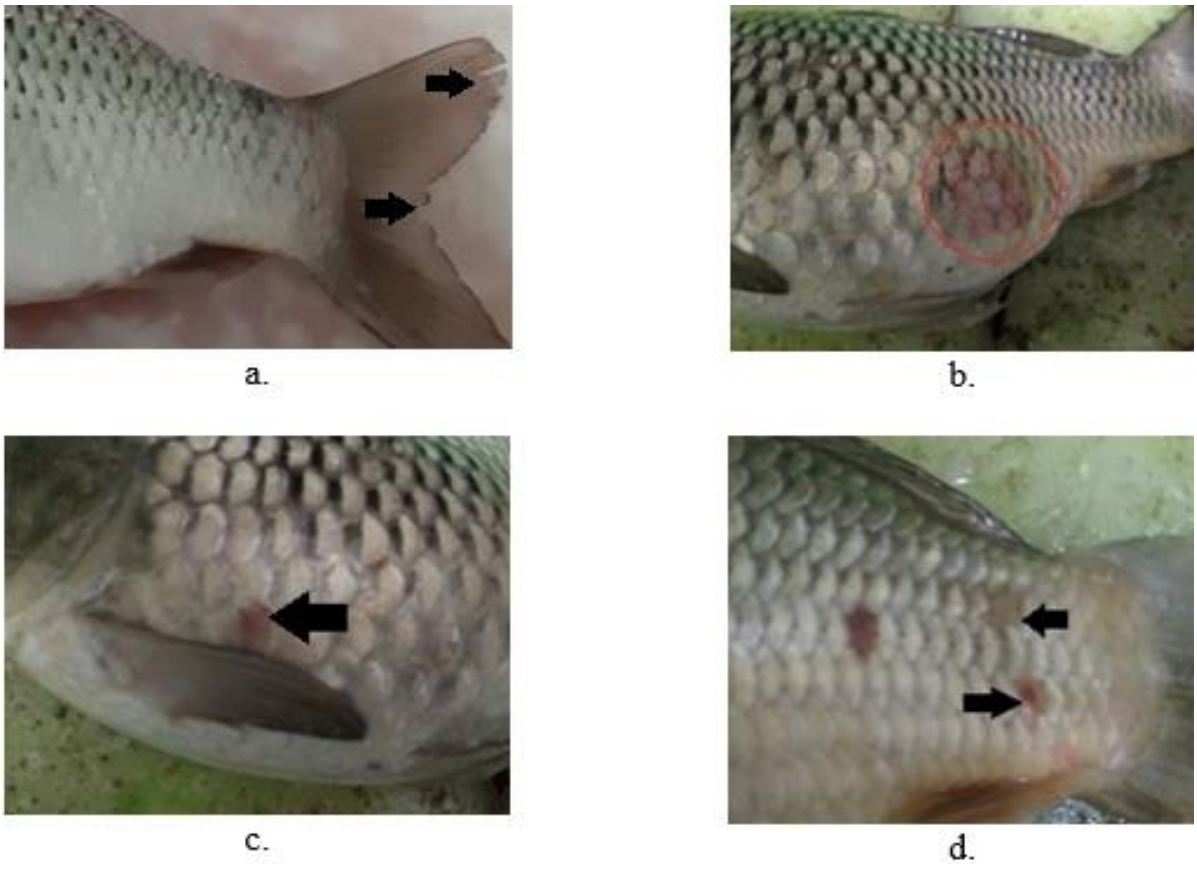

Figure 1. Caudal fin flakes **(a)**, hemorrhage **(b)**, body ulcers **(c)**, and loose scale and body lesions **(d)**.

3. 2. Isolation and Purification

Figure 2. Bacteria growth on TSA after isolation.

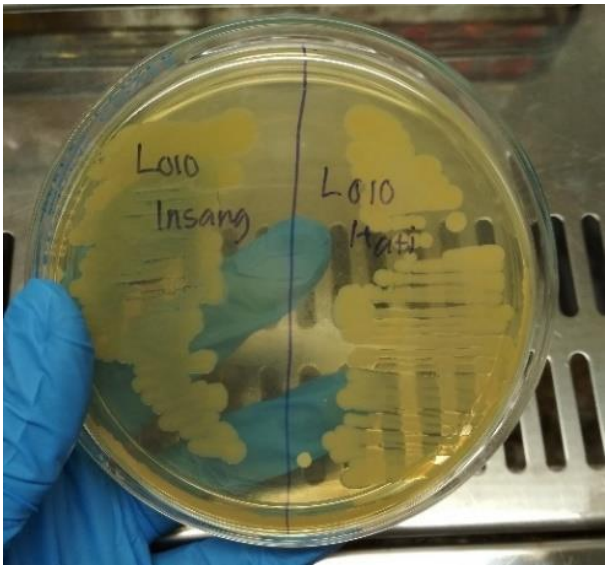

Figure 3. Bacteria growth on TSA after purification.

The results of isolation (**Figure 2**) and bacterial purification (**Figure 3**) of the fish samples showed that the average bacterial colony contained in fish had a yellow to yellow creamy color and a rounded bacterial colony. The color and shape of bacterial colonies that grow on TSA media can help in the process of bacterial identification. Each bacterial colony can have a different shape on the TSA media.

3. 3. KOH String Test, Catalase and Oxidation Test

When KOH string tests are performed, the majority of the bacteria samples produce a string when being pulled up using the inoculation loop. This indicates that most of the bacteria found in the fish samples are gram-negative bacteria. Nonetheless, samples coded L012B and L020B are isolated from skin mucus; samples coded L001A, L010A, L016A, L017A, L018A, L021B, and L022B are isolated from the gills; samples coded L017A, L020B, and L022B are isolated from the liver; also samples coded L010A and L020B are isolated from the kidney are an exception because the string test results showed that they're categorized as gram-positive bacteria for not producing strings when being pulled up using the inoculation loop.

Catalase test results was positive for majority of the samples except for the sample coded L001A, isolated from the gills which is negative. For the Oxidase test, almost the entire samples also are oxidation positive with the exception of samples coded L012B and L020B isolated from the skin mucus; samples coded L001A and L007B isolated from the gills; samples coded L002A, L016A, L018A, and L022B isolated from the liver; and samples coded L018A and L013B isolated from the kidney.

3. 4. Bacteria Identification

Some species of bacteria that have been identified are bacteria that are commonly found in waters and co-exist with other aquatic organisms, such as fish. Bacteria can live on the surface of a fish's body, such as on scales, gills, or even on the internal organs of fish and

symbiotics. But under certain conditions these bacteria can be pathogenic and harmful to fish, that is why some bacteria are categorized as opportunistic pathogenic bacteria [6].

The bacteria that were isolated, the most of them were bacteria from the gram negative group. According to Pekala and Safinska (2018), pathogenic bacteria in fish are generally including gram-negative bacteria. Based on the results of biochemical tests conducted, the most numerous and least genus of bacteria found sequentially were: Aeromonas (37.5%), Plesiomonas (15.6%), Staphylococcus (9.4%), Bacillus (7.8%), Micrococcus (7.8%), Pseudomonas (6.3%), Acinetobacter (4.7%), Citrobacter, Shewanella, Neisseria and Serratia (1.6%). Bacteria were found in every target organs of the fish.

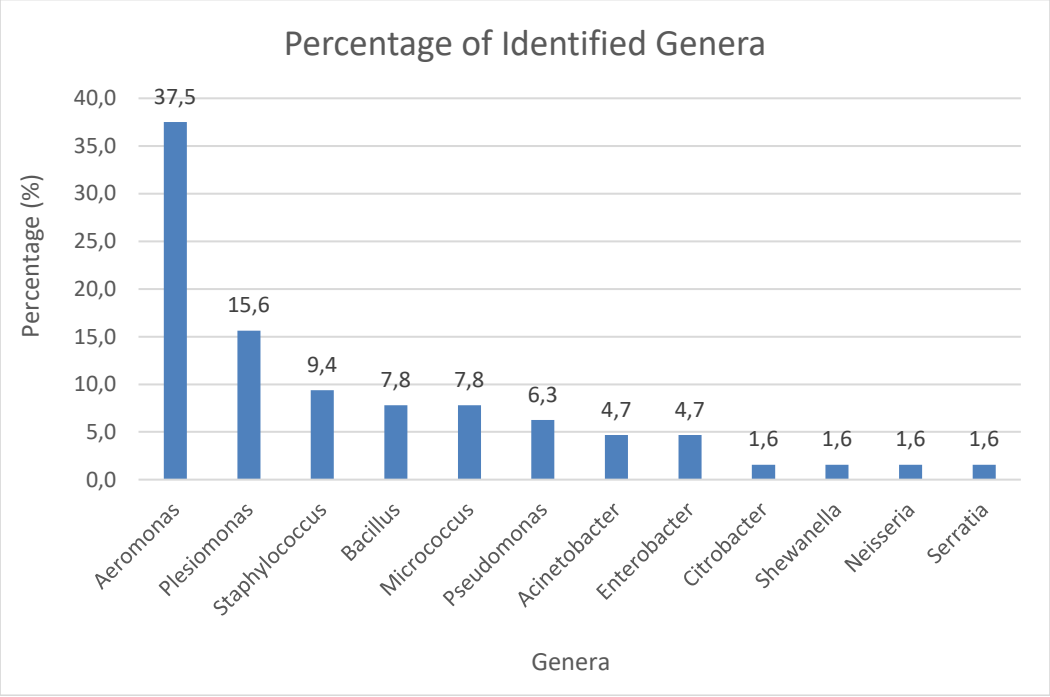

Figure 4. Percentage of identified genera from fish.

3. 5. Aeromonas

Aeromonas bacteria species found were *Aeromonas hydrophila*, *Aeromonas sobria*, *Aeromonas schubertii*, *Aeromonas media*, *Aeromonas caviae* and *Aeromonas eucrenophila*. Aeromonas bacteria were found mostly in isolates from kidney and gill organs, namely as many as seven isolates in each organ. Aeromonas bacteria found in the gills due to gill organs are in direct contact with water. *Aeromonas* spp. is an aquatic microorganism that is commonly found in irrigation waters, rivers, brackish waters, freshwater, estuaries, and saltwater [7]. The biochemical test of Aeromonas all showed the same results on urease, nitrate, and malonate tests which are urease negative, nitrate positive, and malonate negative.

A. hydrophila is a type of bacteria that are opportunistic pathogens and normally live in waters and are ready to strike if the fish conditions are less good and can cause systemic disease which further can lead to death [8]. The decrease in the fish body resistance can be caused by poor water conditions. Water quality is one of the keys to the success of fish farming, so if it

does not meet the requirements, the water will become a source of dangerous diseases [9]. The water contained in the aquariums in both supermarkets is rather turbid in color and there are loose fish scales at the bottom of the aquarium, besides that there is also a buildup of fish droppings in the corner of the aquarium which can cause an increased organic matter content in the water.

Table 1. Aeromonas biochemical test results.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Aeromonas schubertii	+	A/A	F	-	-	+	-	+	+	-	-	+	-
Aeromonas media	-	A/K	F	-	-	+	-	+	+	+	-	+	-
Aeromonas sobria	+	A/A	F	-	-	+	+	+	-	+	-	+	-
Aeromonas hydrophila	+	A/A	F	-	+	+	-	+	+	+	-	+	-
Aeromonas eucrenophila	+	A/K	F	-	-	+	+	-	-	-	-	+	-
Aeromonas caviae	+	A/A	F	-	+	+	+	+	+	-	-	+	-

3. 6. *Plesiomonas shigelloides*

Table 2. *Plesiomonas shigelloides* biochemical test result.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Plesiomonas shigelloides	+	A/K	F	+	+	+	+	+	-	-	+	+	-

Plesiomonas shigelloides are emerging pathogens that are widespread in aquatic environments, the natural distribution of these microorganisms is water and fish in tropical climates [10]. *Plesiomonas* sp. growth in freshwater depends on temperature, nutrient

availability, and the level of contamination of waste in the waters [11]. The presence of Plesiomonas bacteria in aquaria used to sell fish can be caused by the accumulation of organic fish metabolic waste found in the aquarium Another possibility is that these bacteria are bacteria that originate from ponds where fish are raised before being shipped to supermarkets. The biochemical test results of Plesiomonas are shown in **Table 2**.

3. 7. *Staphylococcus epidermidis*

Staphylococcus epidermidis has been reported to be pathogenic in some saltwater and freshwater fish [12]. *Staphylococcus epidermidis* is also a common bacteria found in human epithelial tissue [13], but the Staphylococcus bacteria contained in the water is a different strain to the strain that is commonly found on human skin. This is proven in several studies [14, 15] that have found the bacteria *Staphylococcus epidermidis* in aquatic environments. Staphylococci can stick to fish for a long time without showing signs, disease can arise if there is a sudden increase in temperature or caused by stress [16]. *Staphylococcus epidermidis* is an opportunistic pathogenic bacterium that can be pathogenic in fish when fish are stressed. Eliminating these bacteria from fish or waters is not possible [16]. The biochemical test results of *Staphylococcus epidermidis* are shown in **Table 3**.

Table 3. *Staphylococcus epidermidis* biochemical test result.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Staphylococcus epidermidis	+	A/A	F	-	+	+	+	+	-	+	-	+	-

3. 8. *Micrococcus luteus*

Table 4. *Micrococcus luteus* biochemical test result.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Micrococcus luteus	+	K/K	-	+	-	-	+	-	-	-	-	-	-

M. luteus is a gram-positive bacterium that is pathogenic in fish. The organs that are attacked by these bacteria are the kidney and liver, and they are also found in water [17]. In this research, the bacteria *Micrococcus luteus* were found in the gills, liver and kidneys of fish; bacteria were also found in water samples from the supermarket Transmart. *M. luteus* can usually cause chronic inflammation or infection in adult fish and larval stadia fish [18]. The effect of the pathogenicity of *M. luteus* causes phlegm in certain parts of the body, such as the liver, lymph, and kidney of fish [15]. The biochemical test result of *Micrococcus luteus* is shown in **Table 4**.

3. 9. Pseudomonas

Pseudomonas are found in the liver, kidneys, and mucus on the surface of the fish's body. Pseudomonas species found were *Pseudomonas stutzeri*, *Pseudomonas pseudoalcaligenes*, and *Pseudomonas aeruginosa*. These bacteria are included as Gram- negative bacteria. This bacterium is aerobic, has a short rod, positive catalase, positive oxidase, is unable to ferment but can oxidize glucose / other carbohydrates, is not spherical, has no sheet and has a monotrika flagellum (single flagellum at the poles) so that it always moves [19]. This bacterium belongs to the Pseudomonadaceae family which is a cause of disease in fish and is included as an opportunistic pathogen. The biochemical results of the Pseudomonas bacteria are presented in **Table 5**.

Table 5. Biochemical test result of Pseudomonas.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Pseudomonas stutzeri	+	A/K	F	+	-	+	+	-	-	-	-	+	-
Pseudomonas pseudoalcaligenes	+	K/K	O	+	-	+	-	+	-	-	+	+	-
Pseudomonas aeruginosa	+	K/K	F	-	-	+	+	+	-	-	+	-	-

3. 10. Enterobacter

Enterobacter sp. is a member of the order Enterobacteriales, family Enterobacteriaceae, and genus Enterobacter. In the results of this research, Enterobacter bacteria were found in the liver, kidney, and mucus on the surface of the body. Bacteria of the genus Enterobacter are bacteria that can live in waters and are opportunistic pathogens [20]. Enterobacter bacteria also have antibacterial properties, but also have pathogenic factors including endotoxin and enterotoxin [21]. The results of Enteribacter's biochemical tests are presented in **Table 6**.

Table 6. Biochemical test result of Enterobacter.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Enterobacter aerogenes	+	K/A	F	+	-	+	+	+	-	-	-	+	-
Enterobacter sakazakii	+	A/A	F	-	-	+	+	+	+	-	-	+	-

3. 10. *Citrobacter freundii*

Citrobacter freundii is a species of Enterobacteriaceae, which is often found in water, soil, food, feces and digestive tract in humans and animals. *Citrobacter freundii* can cause lesions of the skin, fins, girls, and internal organs, as well as haemorrhagic and infections. *Citrobacter freundii* in tilapia is showing symptoms of acute septicemia haemorrhage [22]. The results of the biochemical test of *Citrobacter freundii* are presented in **Table 7**.

Table 7. Biochemical test result of *Citrobacter freundii.*

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Citrobacter freundii	+	A/K	F	-	-	+	+	+	+	-	-	+	-

3. 11. *Acinetobacter iwofii*

Acinetobacter lwoffii is often isolated from healthy or diseased fish as part of the bacterial flora of fish because generally bacteria found in the aquatic environment are also present on the skin, gills, and digestive systems of aquatic organisms. Bacteria of the genus Acinetobacter are usually considered to be normal saprophytic organisms. *Acinetobacter lwoffii* is more commonly found in carp (*Cyprinus carpio*) than other Acinetobacter species. The most common symptoms arising in fish infected with the bacterium Acinetobacter are exophthalmia, hemorrhage, and ulcers in the body [23]. Clinical symptoms, such as hemorrhage and the appearance of ulcers on the body were also experienced by fish samples from both supermarkets. The results of the biochemical test of *Acinetobacter lwoffii* is presented in **Table 8**.

Table 8. Biochemical test result of *Acinetobacter iwoffii*.

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Acinetobacter iwofii	+	K/K	O	+	-	+	-	-	-	-	-	+	-

3. 12. *Shewanella putrefaciens*

Shewanella putrefaciens is a bacterium that is commonly found in marine sediments and has been isolated in marine fish. The pathogenicity of this bacterium was first known in marine fish *Siganus rivulatus* [23]. The bacterium *Shewanella putrefaciens* was first isolated from freshwater fish in 2002 in carp (*Cyprinus carpio*) by A. Kozińska and A. Pękala. *S. putrefaciens* is a bacterium that is classified as an opportunistic pathogen and can cause disease in certain conditions, such as stress due to external factors and decreased immunity. Like other opportunistic pathogenic bacteria, *S. putrefaciens* is a physiological flora in saltwater and freshwater fish [24]. In healthy fish, *Shewanella putrefaciens* is usually found in gills [23]. The results of the biochemical test of *Shewanella putrefaciens* is presented in **Table 9**.

Table 9. Biochemical test result of *Shewanella putrefaciens.*

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Shewanella putrefaciens	+	K/K	O	+	-	-	-	-	-	-	-	+	-

3. 13. *Serratia marcescens*

Serratia sp. is a type of gram-negative bacteria, from the family Enterobacteriaceae. This bacterium is in the form of a short stem with a size of 0.5-0.8 × 1.5-5.0 μm. The catalase test is positive, motile, the optimum growth temperature at 30-37 ºC. This bacterium is an anaerobic facultative bacteria that does not really need oxygen [25]. Serratia bacteria are generally a normal flora in the digestive system of fish. *Serratia marcescens* can produce several hydrolytic enzymes, such as protease, chitinase, nuclease, and lipase [26]. The biochemical results of *Serratia mucosa* are presented in **Table 10**.

Table 10. Biochemical test result of *Serratia marcescens.*

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Serratia marcescens	+	A/K	F	-	-	+	-	+	+	+	-	+	-

3. 14. *Neisseria mucosa*

Neisseria mucosa has been identified from animals, waters, and sediments [27]. This proves that these bacteria can live in various hosts and in the host environment. It is not yet known whether *Neisseria mucosa* shows host tropism. Isolation of the bacteria *Neisseria mucosa* in fish itself has not been done much, but because these bacteria can also live in the waters, the possibility of these bacteria to live on the body of the fish is very high. This bacterium is found in mucus on the surface of the body of the sample fish. The biochemical results of *Neisseria mucosa* are presented in **Table 11**.

Table 11. Biochemical test result of *Neisseria mucosa*

Species	Motility	TSIA (Acid/Alkali)	O/F	Ornithin	Indole	Glucose	Citrate (SCA)	Lysin (LIA)	MR	VP	Urease	Nitrate	Malonate
Neisseria mucosa	-	K/K	O	+	-	+	+	+	-	-	-	+	-

4. CONCLUSIONS

Based on the results of research that has been done, the bacterial species isolated and identified from samples of carp obtained from supermarket Transmart and Superindo are: *Aeromonas*hydrophila, *Aeromonas*sobria, *Aeromonas Schubertii*, *Aeromonas media*, *Aeromonas caviae*, *Aeromonas eucrenophila*, *Pseudomonas stutzeri*, *Pseudomonas pseudoalcaligenes*, *Pseudomonas aeruginosa*, *Plesiomonas shigelloides*, *Bacillus megaterium*, *Bacillus thuringiensis*, *Neisseria mucosa*, *Shewanella putrefaciens*, *Citrobacter freundii*, *Serratia marcescens*, *Staphylococcus epidermidis*, *Micrococcus luteus*, *Enterobacter aerogenes*, and *Enterobacter sakazakii*.

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