# THE ISOLATION AND IDENTIFICATION BACTERIA ON JALLALAH ANIMAL (STUDY ON THE FEEDING TILAPIA (*Oreochromis niloticus*) WITH CHICKEN MANURE AS FOODS)

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**Abstract**: In the hadith, Rasulullah *SAW* prohibit human to consume meat and milk from manure-eating animal (*Jallalah* animal). In this study, tilapia fishes were fed chicken manure (being *Jallalah*). Then, it was analyzed based on the microbe in their intestine. The purposes of this study are to isolate and to identify the types of bacteria, and to determine the pathogenic and non-pathogenic bacteria in the intestines of fish that have been fed chicken manure. Bacteria samples were isolated from the intestine of fish with chicken manure and the commercial fish food as a control. Bacteria identification was done by using morphological characterization, macroscopic and microscopic identification, and biochemical test. The results show that 21 isolated bacteria in the fish intestine that was fed chicken manure and six of the bacteria are pathogenic. Based on this study, it can be concluded that tilapia fish by feeding with chicken manure contains many types of pathogens bacteria. Briefly, tilapia fish is not good for being consumption.

**Keywords:** Bacteria; Chicken manure; Jallalah animals; Thoyyib

**Abstrak**: Dalam hadis yang diriwayatkan oleh Ibnu Umar, Rasulullah *SAW*. Melarang memakan daging dan meminum susu hewan pemakan kotoran (hewan *Jallalah*). Pada penelitian ini, ikan nila diberi pakan kotoran ayam (menjadi *Jallalah*). Selanjutnya ikan tersebut dianalisis kandungan mikroba yang terkandung didalam saluran pencernaannya. Tujuan penelitian ini adalah mengisolasi dan mengidentifikasi bentuk bakteri dan mendeterminasi bakteri patogen dan non-patogen yang terdapat didalam saluran pencernaan ikan ynag telah diberi pakan kotoran ayam. Sampel bakteri diisolasi dari saluran pencernaan ikan setelah diberi kotoran ayam dan ikan yang diberi pakan komersil (kontrol). Identifikasi bakteri dilakukan secara makroskopik, mikroskopik, dan uji biokimia. Hasil penelitian menunjukkan ditemukan 21 isolat bakteri dalam usus ikan

yang diberi korotan ayam yang tergolong ke dalam genus Listeria, Staphylococcus, Alcaligenes, Cromobacterium, Edwardisiella, Micrococcus, Bacillus, dan Kurthia. Dari 21 jenis tersebut terdapat 6 jenis bakteri pathogen yaitu *Edwarsiella* sp, *Micrococcus* sp, *Staphylococcus* sp, *Alcaligenes* sp, *Listeria* sp dan *Chromobacterium*. Kesimpulan pada penelitian ini bahwa hewan ikan yang memakan kotoran ayam mengandung banyak jenis bakteri patogen sehingga tidak baik untuk di konsumsi.
**Kata kunci**: Bakteri; hewan *jallalah*; ikan; kotoran ayam; *Thoyyib*

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## Introduction

Halal is a term of lawful or permitted to do according to Islamic law which regulates all aspects of life especially in consuming food (Adawiyah & Kulsum, 2019). Allah SWT commands human not only to consume halal foods but also to consume healthy and clean foods (*halalan thayyiban*). Besides, the important thing that the foods are clean materials from contamination and useful for the human body (Kulsum et al., 2019). Even this command is equated with devotion to Allah, as a very strict and clear command (al-Maidah verse 88), but this command is affirmed in another verse, as found in Surah Al Baqarah: 168.

*"O people! Eat of what is lawful and good on earth, and do not follow the footsteps of Satan. He is to you an open enemy"*.

Hadith above describes that *Jallalah* animals are pretty much narrated by various *mukharrij* with various *sanad*. Based on those various hadith, there are three forms of prohibition, namely eating its eggs, meat and drinking its milk. *Jallalah* animals are forbidden for being human consumption because they eat *najs* such as animal manure even human faeces that certainly contain something harmful for the health beyond any other benefits. Rasullullah hadith about *Jallalah* animals were narrated by Abu Dawud, which means;

"*Has conveyed to us from Usman Abi Syibah, has conveyed to us from Muhammad bin Ishaq from Ibn Ali Najih from Mjahid from Ibn Umar ra he said, 'The messenger of Allah (Rasulullah) forbade the meat (of) Al-Jallalah and drink its milk'* (HR. Abu Dawud)

Chicken manure is one of the wastes produced from the laying hens and broilers. Based on the research of Suryani et al. (2010), chicken manure contains several bacterias named such as *Lactobacillus achidophilus, Lactobacillus reuteri, Leuconostoc mensenteroides* and *Streptococcus thermophilus* which are lactic acid bacteria (BAL). In this study, the feeding used for tilapia is chicken manure which aims to determine the types of bacteria contained in the fish intestines .

Feeding chicken manure into tilapia is based on the principle of assimilation of inorganic nitrogen (ammonia, nitrate, and nitrite) by microbes in the culture medium. Then, it can be used by microbes as the food sources (De Schryver & Verstraete, 2009; Pandey et al., 2014). The contained microbes in the intestines of fish have amylolytic activity, (digesting carbohydrates) proteolytic, (digesting protein), and lipolytic (digesting fat). Based on the study that was conducted by Supriyatna & Ukit (2016), the type of cellulolytic bacteria was found in the intestines of *Hermetia illucens* larvae.

The isolation bacteria is a microbe separation that will be tested from other microbes by using a medium agar. It is expected to obtain a pure culture. Identification bacteria is an activity for determining the carried certain types of organisms that have several steps namely observing, testing, recording, and identifying based on the test results at the fishery. There is often a problem that should be faced. It is the occurrence of disease attacks caused by pathogenic organisms such as bacteria, viruses, fungi, and parasites (Noga, 2010; Woo & Bruno, 2011). Various methods of tackling fish diseases have been done and continued for further development. The general methods used for disease prevention are antibiotics or chemicals, vaccinations, immunostimulants, various medicinal plants and probiotics. The condition of fish is affected by humans' anxiety as the consumers of cultivated products. Because of that, the various safety of disease prevention efforts have to be developed. Therefore we can consume healthy products (Hagi & Hoshino, 2009). Thus the main objectives of this study are to isolate and to identify the bakery which contains *Oreochromis niloticus* at fish intestines which have been fed chicken manure before.

Bacteria identification was done based on morphological characterization, macroscopic and microscopic identification, and biochemical test. Bacteria identification refers to Bergey's Manual of Systemic Bacteriology (Bergey et al., 1984; Brown, 2010) and Cowon and Steel's manual for the Identification of Medical Bacteria (Rahayu et al., 2019).

## Materials and Methods

### Tools and Materials

This research was conducted at the Aquatic Ecology laboratory and Microbiology Laboratory of UIN Sunan Gunung Djati Bandung from January till March 2019. The equipments used were the aquarium which has size 60cm x 50cm x 40cm, aerator, surgical instrument, oven, autoclave, laminar air flow (LAF), balance sheet analytic, beaker glass, vortex, Erlenmeyer, petri dish, test tube, cover glass, pipette, microscope, and measuring cup. The materials used were one month old tilapia fish, chicken manure, distilled water, 70 % alcohol, methylated spirits, physiological NaCl, iodine solution, NA media (nutrient agar), NaCl, crystal violet, safranin, green malachite, 3 % $H_2O_2$ solution, medium TSIA and urea agar-based media.

**Research Procedure**

**An Isolation Bacteria from Tilapia Intestine.**

The fish intestine was taken by using a surgical instrument on Laminar Air Flow (LAF). Then the sample was diluted with 90 ml of physiologically NaCl. The intestinal sample is homogeneous using a vortex. Then the dilution was done from $10^{-1}$ to $10^{-10}$. To obtain $10^{-1}$ dilution. One gram of sample was put into a test tube containing 9 ml of distilled water until we get the $10^{-10}$ dilution. The bacterias' isolation was done by taking one ml of suspension in the dilution series $10^{-3}$ to $10^{-10}$ Then it was isolated by the Spread plate method into the Petri dish aseptically. After that, it was poured into the NA (Nutrient Agar) media and flattened by turning the media in the direction of the eight number to become homogeneous. Bacterias were incubated for 24-48 hours at 37 $^{o}$C. For obtaining the pure isolation, after the colonies grew in each medium, a quadrant stroke technique with several stages was used. The culture was incubated with upside down disk for 24 hours at 35 $^{o}$C (Darmayasa, 2008).

**Identification**

**Macroscopic Identification**

Macroscopic identification includes the size, shape, elevation, edges, and colour of bacteria colonies grew on NA (Nutrient Agar) media. The shapes of the colony are (seen from above) in the form of round (circular), filamentous, irregular, rhizoid, and spindle. Here are some sizes such as pin point, large, medium, and small. The colony surfaces/elevation (seen from the side) are flat, raised, convex, and umbonate. The edges of the colony (seen from above) are entire, lobate, serrate, filamentous, and undulate. The colony colours are whitish, grey, yellowish or almost clear (Dwidjoseputro, 1989).

**Microscopic Identification**

**The staining gramme**

The isolated bacterias were put on the object glass and were dropped with staining reagent (Gentian Violet). Also, they were kept for one minute. Then, it washed with flowing water and be dried. After that, they were dropped with iodine and alcohol for one minute. The isolated bacteria on object glass were observed with objective 1000x magnification. Under the microscope, the gram of positive bacteria will appear into the purple one and the negative will appear into the red one (Mohamad et al., 2014)

**Endospore test**

Endospore test is done in order to determine the evolution responding of the bacteria of the environment. It is the most resistant transformation. It can survive in unfavourable environments. Endospore resistance is due to the thick

spore sheath. The isolated bacterias were dropped with the malachite green of staining reagent for ten minutes. Then they were dropped and left with safranin solution for one minute. After that, they were observed under a microscope with strong magnification. The presence of endospores was indicated with green colour, and parts of cells that do not contain endospores were indicated with bright red colour (Mohamad et al., 2014).

**Motility test**

The isolated bacterias were inserted into semi-solid NA media by stab method by using a sterile ose needle. Then they were incubated for 24 hours at 37$^{o}$C. The positive results were marked by the growth of bacteria spread throughout the medium, and the negative results were characterized by the growth of shrink bacteria (Maulani et al., 2019).

**Biochemical Test**

**TSIA**

The bacteria colony was taken by using ose and inoculated on TSIA media by stabbing through the centre of the medium to the bottom (butt) of the tube. Then it was taken by streaking zigzag on the surface of the slant. After that, the tube was incubated at 37 ºC for 24 hours by observing the medium of colour changes. If the slant is red and the butt is yellow, it will cause the bacterias are able to ferment glucose. Whereas if both of the slant and butt are yellow, the bacteria are able to ferment sucrose and lactose (Abdallah et al., 2018).

**Catalase test**

As much as one drop of 3 % $H_2O_2$, it was dropped on a glass object. Then the isolated bacterias were applied and mixed slowly by using an ose needle. Then, the positive results are marked by the formation of air bubbles in the surrounding colonies (Begum et al., 2017).

**$H_2S$ Test**

The bacteria colony was inoculated into TSIA media. Stubbing to the butt and streaking the slant, it was incubated at 37 $^{o}$C for 24 hours. Positive reactions are characterized by the presence of black precipitate on media (Brown, 2010; Begum et al., 2017).

**Urease Test**

The isolated bacterias were inoculated on the surface of urea media and incubated for 24 hours at 29 ºC. The positive result was marked by the colour changes of the medium become blue and the negative result was marked by the unchanged colour of the media (Begum et al., 2017).

## Results and Discussion

The isolation and Identification of the isolated bacteria from the intestines of the feeding tilapia with chicken manure (*Jallalah*)

### Macroscopic Observation of Bacteria on Feeding Tilapia with Chicken Manure (*Jallalah*)

Macroscopic observation of bacteria was done by looking at bacteria morphology based on the size, shape, elevation, edges, colour, and colony surface (Figure 1).

**Figure 1.** Macroscopic observation of bacteria with Dilution $10^{-6}$

The first figure represents the purification process of isolated bacteria at dilution $10^{-6}$. After the purification, the bacterias were rejuvenated (cultivation). Bacteria cultivations were done to obtain a culture supply. Cultivation was made triple as inventory to replace contaminated isolation during the observation. The following step is the macroscopic observation of isolated bacteria from the intestines of the feeding tilapia with chicken manure (Jallalah) (Tabel 1) and bacteria in intestines of the feeding tilapia with the commercial fish (Table 2).

**Table 1.** The macroscopic test result of bacteria morphological characteristic (three samples) of intestinal tilapia manure

| Code | Macroscopic | | | | | |
|---|---|---|---|---|---|---|
| | **Size** | **Type** | **Elevation** | **Side** | **Colour** | **Surface** |
| IS-KA1 | Small | Irregular | Flat | Lobate | White | Smooth |
| IS-KA2 | Small | Circular | Flat | Entire | White | Smooth |
| IS-KA3 | Pin point | Circular | Flat | Entire | White | Smooth |
| IS-KA4 | Small | Irregular | Flat | Lobate | White | Smooth |
| IS-KA5 | Small | Irregular | Flat | Entire | Cream | Smooth |
| IS-KA6 | Pinpoint | Circular | Flat | Entire | Yellow | Smooth |

| Code | Macroscopic | | | | | |
|---|---|---|---|---|---|---|
| | **Size** | **Type** | **Elevation** | **Side** | **Colour** | **Surface** |
| IS-KA7 | Pinpoint | Circular | Flat | Entire | White | Smooth |
| IS-KA8 | Small | Irregular | Flat | Lobate | White | Smooth |
| IS-KA9 | Small | Circular | Flat | Entire | White | Smooth |
| IS-KA10 | Small | Irregular | Flat | Undulate | White | Smooth |
| IS-KA11 | Small | Circular | Convex | Entire | Violet | Smooth |
| IS-KA12 | Small | Circular | Umbonate | Entire | White | Smooth |
| IS-KA13 | Large | Irregular | Flat | Lobate | White | Smooth |
| IS-KA14 | Small | Irregular | Flat | Lobate | White | Smooth |
| IS-KA15 | Small | Circular | Flat | Entire | White | Smooth |
| IS-KA16 | Small | Irregular | Flat | Lobate | White | Smooth |
| IS-KA17 | Small | Circular | Flat | Entire | White | Smooth |
| IS-KA18 | Middle | Irregular | Flat | Undulate | White | Smooth |
| IS-KA19 | Small | Circular | Flat | Entire | White | Smooth |
| IS-KA20 | Pinpoint | Circular | Flat | Entire | White | Smooth |
| IS-KA21 | Small | Circular | Flat | Entire | White | Smooth |

**Table 2.** Results of macroscopic observations of bacteria morphology in the intestines of fish fed commercial feed

| Code | Morphological Characteristic | | | | | |
|---|---|---|---|---|---|---|
| | **Size** | **Type** | **Elevation** | **Side** | **Colour** | **Surface** |
| IS-K1 | Small | Irregular | Flat | Lobate | W | SM |
| IS-K2 | Pinpoint | Circular | Convex | Entire | W | SM |
| IS-K3 | Small | Irregular | Flat | Lobate | W | SM |
| IS-K4 | PP | Circular | Convex | Entire | W | SM |
| IS-K5 | Large | Filamentous | Flat | Lobate | W | SM |

Based on table 1, the intestine of tilapia fish that have been fed chicken manure contained 21 isolated bacterias, while there are only five isolated bacteria that were successfully isolated from the intestines of fish from the commercial foods. (Table 2). The difference number of isolated bacteria and morphological characteristic was influenced by different feeding. The types of bacteria in the fish intestine that was feeding commercial foods (control) are probiotic bacteria that play a role in the growth of fish. This is appropriate with Dar et al. (2016a), Dar et al. (2016b) in his research on bacteria isolation in fish that eat faeces. There were found pathogenic bacterias namely *Escherichia coli* and *Staphylococcus faecalis*. Furthermore, to find out the genus of bacteria, microscopic observations were obtained.

## Microscopic Observation

The Microscopic observations were done based on cell shape, gram, the presence of endospore, and mortality. The following table shows the result of microscopic isolation analysis of bacteria.

**Table 3.** Microscopic observations of bacteria morphology in the intestines of fish fed chicken manure (Jallalah)

| Code | Morphological Character | | | |
|---|---|---|---|---|
| | **Type** | **Gram** | **Endospore** | **Motility** |
| IS-KA1 | Bacillus | + | - | + |
| IS-KA2 | Bacillus | - | - | + |
| IS-KA3 | Coccus | + | - | + |
| IS-KA4 | Bacillus | - | - | + |
| IS-KA5 | Coccus | + | - | + |
| IS-KA6 | Coccus | + | - | + |
| IS-KA7 | Bacillus | + | + | + |
| IS-KA8 | Bacillus | + | + | + |
| IS-KA9 | Coccus | + | - | + |
| IS-KA10 | Bacillus | - | - | + |
| IS-KA11 | Bacillus | - | - | + |
| IS-KA12 | Bacillus | + | - | + |
| IS-KA13 | Bacillus | + | + | + |
| IS-KA14 | Bacillus | + | + | + |
| IS-KA15 | Bacillus | + | - | + |
| IS-KA16 | Bacillus | + | - | + |
| IS-KA17 | Coccus | + | - | + |
| IS-KA18 | Bacillus | + | + | + |
| IS-KA19 | Bacillus | - | - | + |
| IS-KA20 | Bacillus | + | - | + |
| IS-KA21 | Coccus | + | - | + |

Based on microscopic observation, there were 15 isolated bacteria with bacillus shapes and six isolated bacteria with coccus shapes. Based on the gram characteristic, there are 16 gram positive of isolated bacteria that can maintain the purple colour (violet crystals) and five gram isolated bacterias are negative which marked with red cells. This is appropriate with Morrison & Rubin (2020) which state bacteria that absorb Gram A (violet crystals) will remain purple after fading with Gram C (Alcohol acetone). It is called Gram Positive Bacteria. bacteria whose colour is faded in washing with alcohol. It will absorb Gram D (Safranin) stain. The pink one is called Gram negative bacteria.

Gram-negative bacterias have cell walls with high lipid content in the form of lipopolysaccharides and lipoproteins. During the colouring process, the lipids will dissolve by alcohol. It caused the colour of the violet crystals in the cell wall cannot be maintained and will bind to the red colour derived from safranin. On

gram-positive bacteria, the cell wall consists of peptidoglycan. It is not capable of being dissolved by acetone alcohol. It caused the purple colour of violet crystals can be maintained. Gram-positive bacterias have 40 sheets of peptidoglycan layers which make up to 50 % of the total cell wall material. While gram-negative bacteria only have one or two sheets of peptidoglycan layers, which means only 5-10 % of the total cell wall as the material (Jawetz et al., 2010).

Based on the presence of endosposres, five isolated bacterias have endospores. However, 16 isolated bacteria do not have endospores. The existence of endospore was seen from the presence of green bacteria cells among other red cells. According to Laue et al. (2018) bacteria, endospores structures are resistant for being extreme environmental conditions such as dry, heating, and acidic conditions.

The result of motality test shows that 20 isolated bacterias with positive reactions and one isolated bacteria with negative reaction. Positive results are indicated by the presence of white colour around the stab area and the formation of a white ring on the surface of the media. This is according to Waluyo (2008), motile positive isolated bacterias are shown by spreading the isolated bacyerialto all media surfaces.

Motility is a test which was done by determining whether there is the movement of a microorganism through flagella or not. According to Creppy et al. (2019), the ability of the organism movement is called motility. Almost all bacillus bacterias are motile, whereas the bacteria that form coccus are immotile.

**Table 4.** Microscopic observations of bacteria morphology in the intestines of fish fed commercial feed

| Code | Morphological Characteristic | | | |
|---|---|---|---|---|
| | **Type** | **Gram** | **Endospore** | **Motility** |
| IS-K1 | Bacillus | + | + | + |
| IS-K2 | Bacillus | + | + | + |
| IS-K3 | Bacillus | + | - | + |
| IS-K4 | Bacillus | + | + | + |
| IS-K5 | Bacillus | + | - | + |

Based on table 4 regarding the microscopic morphology of bacteria from the intestines of tilapia with commercial foods, there are three isolated bacteria with bacillus shape and two isolated bacteria with round (coccus) shape. The gram character of the five isolated bacteria was positive. There were three isolated bacterias which produced endospores and two of them does not produce endospores. The motility test shows that all the isolated bacterias were obtained motile which was marked by the formation of white coloured rings on the surface of the media.

The existence of endospore was seen from the presence of green bacteria cells among other red cells. According to Laue *et al*. (2018) bacteria, endospores are structures that resistant to extreme environmental conditions such as dryness, heating, and acidity. Motility is a carried test to determine whether there is the movement of a microorganism through *flagella* or not.

According to Creppy et al. (2019), Karimi et al. (2013) the ability of the movement organism is called motility. Almost all bacillus bacterias are motile, whereas the coccus bacteria are immotile.

## Biochemical Test

Biochemical testing aims to identify and to determine a pure bacteria culture that was isolated based on a physiological characteristic (Begum et al., 2017). Biochemical tests (Table 5) included catalase, glucose, sucrose, lactose tests use TSIA (Triple Sugar Iron Agar) media. however, gas formation tests, $H_2S$ formation, and urease tests use UAB (Urea Agar Base) media.

**Table 5.** Biochemical test results of bacteria isolates from the intestine of tilapia fed commercial feed (TSIA Test: Glu = glucose, Suc = sucrose, Lac = lactose)

| Isolate Code | Biochemical Test | | | | | | |
|---|---|---|---|---|---|---|---|
| | Catalase | TSIA | | | | | Urease |
| | | Glu | Suc | Lac | Gas | $H_2S$ | |
| IS-KA1 | + | + | + | + | - | - | - |
| IS-KA2 | + | + | + | + | - | - | - |
| IS-KA3 | + | + | + | + | - | - | + |
| IS-KA4 | - | + | - | - | + | + | - |
| IS-KA5 | + | + | + | + | - | - | - |
| IS-KA6 | + | + | + | + | - | - | + |
| IS-KA7 | - | + | + | + | - | - | + |
| IS-KA8 | - | + | - | - | + | - | + |
| IS-KA9 | + | + | + | + | + | - | + |
| IS-KA10 | + | + | + | + | + | - | - |
| IS-KA11 | - | + | - | - | - | - | - |
| IS-KA12 | + | + | - | - | - | - | + |
| IS-KA13 | + | + | + | + | + | - | + |
| IS-KA14 | + | + | - | - | - | - | + |
| IS-KA15 | + | + | - | - | - | - | - |
| IS-KA16 | + | - | - | - | - | - | + |
| IS-KA17 | + | + | - | - | - | - | + |
| IS-KA18 | + | - | - | - | - | - | + |
| IS-KA19 | + | - | - | - | - | - | + |
| IS-KA20 | + | + | - | - | - | - | + |
| IS-KA21 | + | + | - | - | - | - | + |

Based on macroscopic and microscopic observation, and biochemical tests on isolated bacteria from intestinal tilapia that has been fed chicken manure

(*jallalah*), obtained nine bacteria genera namely *Listeria*, *Staphylococcus*, *Alcaligenes*, *Amphybacillus*, *Chromobacterium*, *Edwardisiella*, *Micrococcus*, *Bacillus* and *Kurthia* (Table 6).

**Table 6.** Identified genus of bacteria from Tilapia intestine fed chicken manure (Jallalah)

| Isolate Code | Bacteria Genus |
|---|---|
| IS-KA1 | *Listeria* sp. |
| IS-KA2 | *Alcaligees* sp. |
| IS-KA3 | *Staphylococcus* sp. |
| IS-KA4 | *Edwardsiella* sp. |
| IS-KA5 | *Staphylococcus* sp. |
| IS-KA6 | *Micrococcus* sp. |
| IS-KA7 | *Bacillus* sp. |
| IS-KA8 | *Amphybacillus* sp. |
| IS-KA9 | *Staphylococcus* sp. |
| IS-KA10 | *Alcaligees* sp. |
| IS-KA11 | *Cromobacterium* sp. |
| IS-KA12 | *Kurthia* sp. |
| IS-KA13 | *Bacillus* sp. |
| IS-KA14 | *Bacillus* sp. |
| IS-KA15 | *Kurthia* sp. |
| IS-KA16 | *Kurthia* sp. |
| IS-KA17 | *Micrococcus* sp. |
| IS-KA18 | *Bacillus* sp. |
| IS-KA19 | *Alcaligees* sp. |
| **IS-KA20** | ***Kurthia* sp.** |
| IS-KA21 | *Micrococcus* sp. |

Based on the table above, 21 isolated bacteria of purified bacteria divided into nine different genera of bacteria. They are one isolated bacteria from genus *Listeria*, three isolated bacteria from genera *Alcaligenes*, one isolated bacteria belong to genus *Chromobacterium*, three isolated bacteria from genera *Staphylococcus*, one isolated bacteria from genus *Edwardsiella*, three isolated bacteria from genus *Micrococcus*, three isolated bacteria from genus *Bacillus*, one isolated bacteria from genus *Amphybacillus*, and five isolated bacteria belong to genus *Kurthia* (Table 6). From those nine genera, there are six pathogenic bacteria genera namely *Listeria*, *Staphylococcus*, *Alcaligenes*, *Edwardsiella*, *Micrococcus*, and *Kurthia*. Earlier in the study of Aziz (2012) there also were found *Staphylococcus*, *Bacillus*, and *Edwardsiella* bacteria isolated from the digestive tract of tilapia. The genus *Alcaligenes*, *Amphibacillus* and *Micrococcus* were also found on research from Mulia et al. (2011). The genus *Chromobacterium* has been found in the digestive tract of eel fish (Wahyuniati, 2014). The genus Listeria was also discovered by (Novianti, 2012) in the study on identification of the bacteria digestive tract of an indo-pacific bottlenose dolphin (*Tursiops aduncus*).

**Table 7.** Identified genus of bacteria from Tilapia intestine fed commercial feed

| Isolate Code | Bacteria Genus |
|---|---|
| IS-K1 | *Bacillus* sp. |
| IS-K2 | *Bacillus* sp. |
| IS-K3 | *Micrococcus* sp. |
| IS-K4 | *Bacillus* sp. |
| IS-K5 | *Micrococcus* sp. |

Based on the identification of intestinal bacteria of tilapia commercial foods, there are from purification which are divided into two genera of bacteria, three from genera *Bacillus* and 2 from genera *Micrococcus*. The five types of bacteria are probiotics that help of the growth process (Table 7).

The type of bacteria in tilapia intestine as the commercial food is less than the type of bacteria in the intestines of the feeding tilapia with chicken manure as foods. Also, the dominated bacteria were obtained by non-pathogenic bacteria, namely *Bacillus* sp. It is one of the probiotic bacterias that can suppress the growth of pathogenic bacteria. According to LeBlanc et al. (2017), Rattanachaikunsopon & Phumkhachorn (2010), Audrain et al. (2015), the produced compounds in the metabolism of probiotic bacteria such as lactic acid, hydrogen peroxide, bacteriocin are antimicrobial and antibiotic. It is to suppress or inhibit the growth of pathogenic bacteria through the production of antimicrobial compounds. They improve microbial balance in the digestive tract, provide a positive influence of the physiology and health, stimulate the immune system and improve water quality.

## Conclusion

There are 21 isolated bacteria with different characteristics isolated from intestinal the feeding tilapia with chicken manure as Jalallah fish which are divided into nine genera of bacteria namely *Listeria, Alcaligenes, Staphylococcus, Amphybacillus, Micrococcus, Chromobacterim,* and *Edwardisiella. Bacillus*, and *Kurthia.* There are six genera of pathogenic isolated bacteria from the intestines of the feeding tilapia (*Oreochromis niloticus*) with chicken manure as Jalallah fish namely *Listeria, Alcaligenes, Staphylococcus, Micrococcus, Chromobacterim,* and *Edwardisiella*.

## References

Abdallah, M. S., Mustapha, T., Gambo, A., & Ishaq, S. (2018). Biochemical Identification and Cultural Characterization of Some Gram- Negative Bacteria Obtained From Fecal / Diarrhoeal Biochemical Identification and Cultural Characterization of Some Gram- Negative Bacteria Obtained From Fecal / Diarrhoeal Samples. *CIBTech Journal of Microbiology*, *5*(1), 31–34.

Adawiyah, A., & Kulsum, Y. (2019). Study of Critical Point Analysis on Meat-Based Foods in Bandung. *Indonesian Journal of Halal Research*, *1*(2), 40–45. https://doi.org/10.15575/ijhar.v1i2.5780

Audrain, B., Farag, M. A., Ryu, C. M., & Ghigo, J. M. (2015). Role of bacterial volatile compounds in bacterial biology. *FEMS Microbiology Reviews*, *39*(2), 222–233. https://doi.org/10.1093/femsre/fuu013

Begum, K., Mannan, S. J., Rezwan, R., Rahman, M. M., Rahman, M. S., & Nur-E-Kamal, A. (2017). Isolation and characterization of bacteria with biochemical and pharmacological importance from soil samples of Dhaka city. *Dhaka University Journal of Pharmaceutical Sciences*, *16*(1), 129–136. https://doi.org/10.3329/dujps.v16i1.33390

Bergey, D. H., Krieg, N. R., & Holt, J. G. (1984). *Bergey's manual of systematic bacteriology*. Williams & Wilkins.

Brown, L. R. (2010). Microbial enhanced oil recovery (MEOR). *Current Opinion in Microbiology*, *13*(3), 316–320. https://doi.org/https://doi.org/10.1016/j.mib.2010.01.011

Creppy, A., Clément, E., Douarche, C., D'Angelo, M. V., & Auradou, H. (2019). Effect of motility on the transport of bacteria populations through a porous medium. *Phys. Rev. Fluids*, *4*(1), 13102. https://doi.org/10.1103/PhysRevFluids.4.013102

Dar, G. H., Dar, S. A., Kamili, A. N., Chishti, M. Z., & Ahmad, F. (2016). Detection and characterization of potentially pathogenic Aeromonas sobria isolated from fish Hypophthalmichthys molitrix (Cypriniformes: Cyprinidae). *Microbial Pathogenesis*, *91*, 136—140. https://doi.org/10.1016/j.micpath.2015.10.017

Dar, G. H., Kamili, A. N., Chisthi, M. Z., Dar, S. A., Tantry, T. A., & Ahmad, F. (2016). Characterization of Aeromonas sobria Isolated from Fish Rohu (Labeo rohita) Collected from Polluted Pond. *Journal of Bacteriology & Parasitology*, *7*(3), 3–6. https://doi.org/10.4172/2155-9597.1000273

Darmayasa, I. B. G. (2008). Isolasi dan Identifikasi Bakteri Pendegradasi Lipid (Lemak) Pada Beberapa Tempat Pembuangan Limbah dan Estuari DAM Denpasar. *Jurnal Bumi Lestari*, *8*(2), 122–127.

https://doi.org/10.1017/CBO9781107415324.004

De Schryver, P., & Verstraete, W. (2009). Nitrogen removal from aquaculture pond water by heterotrophic nitrogen assimilation in lab-scale sequencing batch reactors. *Bioresource Technology*, *100*(3), 1162–1167. https://doi.org/10.1016/j.biortech.2008.08.043

Dwidjoseputro, D. (1989). *Dasar-Dasar Mikrobiologi*. Djambatan.

Hagi, T., & Hoshino, T. (2009). Screening and Characterization of Potential Probiotic Lactic Acid Bacteria from Cultured Common Carp Intestine. *Bioscience, Biotechnology, and Biochemistry*, *73*(7), 1479–1483. https://doi.org/10.1271/bbb.80746

Jawetz, E., Melnick, E. A., & Adelberg. (2010). *Mikrobiologi Kedokteran*. Salemba Medika.

Karimi, A., Yazdi, S., & Ardekani, A. M. (2013). Hydrodynamic mechanisms of cell and particle trapping in microfluidics. *Biomicrofluidics*, *7*(2), 1–23. https://doi.org/10.1063/1.4799787

Kulsum, Y., Adawiyah, A., Shofwaturrohmani, F., & Nurjanah, D. (2019). Pig Sample Handling in Research Laboratory Scale. *Indonesian Journal of Halal Research*, *1*(1), 14–17. https://doi.org/10.15575/ijhar.v1i1.4091

Laue, M., Han, H. M., Dittmann, C., & Setlow, P. (2018). Intracellular membranes of bacterial endospores are reservoirs for spore core membrane expansion during spore germination. *Scientific Reports*, *8*(1), 1–12. https://doi.org/10.1038/s41598-018-29879-5

LeBlanc, J. G., Chain, F., Martín, R., Bermúdez-Humarán, L. G., Courau, S., & Langella, P. (2017). Beneficial effects on host energy metabolism of short-chain fatty acids and vitamins produced by commensal and probiotic bacteria. *Microbial Cell Factories*, *16*(1), 1–10. https://doi.org/10.1186/s12934-017-0691-z

Maulani, B. I. G., Rasmi, D. A. C., & Zulkifli, L. (2019). Isolation and characterization of endophytic bacteria from mangrove Rhizophora mucronata Lam. and antibacterial activity test against some pathogenic bacteria. *Journal of Physics: Conference Series*, *1402*(3), 1–7. https://doi.org/10.1088/1742-6596/1402/3/033038

Mohamad, N. A., Jusoh, N. A., Zaw Htike, Z., & Lei Win, S. (2014). Bacteria Identification From Microscopic Morphology: A Survey. *International Journal on Soft Computing, Artificial Intelligence and Applications*, *3*(2), 1–12. https://doi.org/10.5121/ijscai.2014.3201

Morrison, B. J., & Rubin, J. E. (2020). Detection of multidrug-resistant Gram-

negative bacteria from imported reptile and amphibian meats. *Journal of Applied Microbiology*, *129*(4), 1053–1061. https://doi.org/https://doi.org/10.1111/jam.14658

Mulia, D. S., Maryanto, H., & Purbomartono, C. (2011). Isolasi, Karakterisasi, dan Identifikasi Bakteri Pada Lele Dumbo Yang Terserang Penyakit di Kabupaten Banyumas. *Sainteks*, *7*(1), 1–15.

Noga, E. J. (2010). *Fish Disease : Diagnosis and Treatment* (2nd ed.). John Wiley and Sons, Inc. USA.

Novianti, M. I. (2012). Identifikasi Bakteri Saluran Pencernaan Lumba-Lumba Hidung Botol Indo-Pasifik (Tursiops aduncus). In *Fakultas Kedokteran Hewan Institut Pertanian Bogor*. Institut Pertanian Bogor.

Pandey, P. K., Kass, P. H., Soupir, M. L., Biswas, S., & Singh, V. P. (2014). Contamination of water resources by pathogenic bacteria. *AMB Express*, *4*(1), 1–16. https://doi.org/10.1186/s13568-014-0051-x

Rahayu, Y. S., Yuliani, & Trimulyono, G. (2019). Isolation and identification of hydrocarbon degradation bacteria and phosphate solubilizing bacteria in oil contaminated soil in Bojonegoro, East Java, Indonesia. *Indonesian Journal of Science and Technology*, *4*(1), 134–147. https://doi.org/10.17509/ijost.v4i1.14923

Rattanachaikunsopon, P., & Phumkhachorn, P. (2010). Lactic acid bacteria: their antimicrobial compounds and their uses in food production. *Annals of Biological Research*, *1*(4), 218–228.

Supriyatna, A., & Ukit. (2016). Screening and Isolation of Cellulolytic Bacteria from Gut of Black Soldier Flays Larvae (Hermetia illucens) Feeding with Rice Straw. *Biosaintifika: Journal of Biology & Biology Education*, *8*(3), 314–320. https://doi.org/10.15294/biosaintifika.v8i3.6762

Suryani, Y., Astuti, Oktavia, B., & Umniyati, S. (2010). Isolasi dan Karakteristik Bakteri Asam Laktat Dari Limbah Kotoran Ayam Sebagai Agensi Probiotik dan Enzim Kolestrerol Reduktase. *Prosiding Seminar Nasional Biologi Yogyakarta*, 138–147.

Wahyuniati, I. (2014). *Bacteria and parasitic worms in the liver and digestive tract of eel (Monopterus albus)*. Institut Pertanian Bogor.

Waluyo, L. (2008). *Teknik dan Metode Dasar dalam Mikrobiologi*. UMM Press.

Woo, P. T. K., & Bruno, D. (2011). *Fish Disease and Disorders : Viral, Bacteria, and Fungial Infections Vol. 3* (2nd ed.). CAB International.