Description of Haematology and Histopathology of Duck Digestive System With UV Light Irradiated *Salmonella* sp

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Abstract. *Salmonella* sp is a cause of Salmonellosis in ducks. One way to kill these bacteria is to use ultraviolet (UV) light. In this study irradiation for 15, 25 and 35 minutes. *Salmonella* sp bacteria can attack several physiological systems of the body, one of which is the digestive system. This is because bacteria infect ducks through food and drink that has been contaminated with bacteria. To see the effect of Salmonellosis, a blood sample and organ of the ducks were removed every 2 weeks from the treatment. The physiological condition of ducks infected with *Salmonella* sp can be seen in hematological conditions and histopathological picture of the liver and intestines in ducks that have been exposed to ultraviolet. Based on the results of research that has been done, it was found that the illumination of *Salmonella* sp using UV light for 35 minutes was most effective to weaken the bacteria because the results showed the smallest value compared to other controls and irradiation. Some histological abnormalities were found in the liver and intestines but still in a condition of minor damage because the damage was only found a little. Abnormalities that occur in the liver include bleeding, nucleus picnosis, necrosis and inflammation, while in the intestine found abnormalities include desquamation of epithelial cells, bleeding and proliferation of goblet cells.

1. Introduction
One of the diseases that mostly attacks ducks is Salmonellosis caused by *Salmonella* sp. This bacterium can suppress the immune system of poultry (immune suppression) and cause the death of broilers [1]. Young ducks will be easily infected with bacteria and if this duck can live to adulthood it will act as a carrier or carrier and can produce eggs that are infected with *Salmonella* sp. Bacterial infections can be transmitted to humans through eggs if the egg processing is not cooked properly, other than that the bacteria can be transferred to newly hatched ducks.

One way to kill bacteria is to use ultraviolet (UV) light. The use of UV light is the best choice for killing bacteria without causing adverse effects on the environment. UV rays are effective in killing microorganisms, viruses and protozoa [2]. The results of the study Ref.[3] stated that irradiation using Ultra Violet rays for 25 minutes with a spacing of 10 cm indicates the bacteria was dead. This is indicated by the absence of bacteria that grow on culture media. With the results of the 25 minute irradiation, *Salmonella* sp is expected to be able to function as a vaccine and be able to increase the immune system in the body. Ref. [4] research results show that making a vaccine with ultraviolet irradiation can produce the best antisera titre at irradiation of 320 nm for 8 x irradiation for each 5 minute exposure. This
bacterium causes diseases of the digestive organs and invasion of blood vessels. Salmonellosis sufferers are usually caused due to decreased immunity and poor hygiene of food consumed.

Salmonella sp can cause fever which is an acute infectious disease in the form of bleeding, liver damage, small intestinal infection and meningitis. During an infection, the bacteria will multiply in phagocytic mononuclear cells and be continuously released into the bloodstream. In patients with Salmonellosis sometimes also found enlarged liver and spleen accompanied by pain [5]. Based on the results of research that has been done, macroscopic examination of the liver in bacterial infections shows the presence of hepatomegaly, namely the liver becomes hyperemic, soft, yellowish and slightly enlarged. Meanwhile, microscopic examination shows that there is inflammation in the portal area, hydropic degeneration, vacuolization, the nucleus becomes karyorexis and finally becomes karyolysis [6]. This research aimed to see the effect radiation of Salmonella sp using UV light for 35 minutes in histological abnormalities in the liver and intestines.

2. Material and Method
The research was carried out at the Microbiology Laboratory, Animal Physiology Laboratory, Biology Study Program, FMIPA, Udayana University and the Great Veterinary Center (BBVet) Denpasar. The study was conducted from June to August 2019.

2.1. Tools and materials
The materials used were 20 3-day-old ducklings that would be divided into 5 groups (K Control, P0 without irradiation, P1 Irradiation 10 minutes, P2 Irradiation 25 minutes and P3 Irradiation 35 minutes), Culture of Salmonella sp bacteria, Lactose Broth media (Lactose Broth media) LB), Nutrient Broth (NB) media, SSA (Salmonella Shigella Agar) media, distilled water, Buffered Neutral Formalin (BNF) 10%, Alcohol 70%. Tools used include cotton buds, test tubes, petri dishes, preparation tools, Auto hematology Analyzer RT 7600

2.2. Work procedures.
The work procedure in this study was divided into 3 stages:

2.2.1. Isolation of Salmonella sp
The pathogenic Salmonella sp is isolated from the cloaca of adult ducks that are indicated as having Salmonellosis. Ducks are taken randomly from duck breeders in the Kerobokan, Badung area. The duck cloaca is swabbed using a sterile cotton bud and then the cotton bud is put into a test tube that contains the Lactose Broth (LB) media, then incubated for 24 hours. After 24 hours, the samples were planted in SSA media and re-incubated for 24 hours. Furthermore, it is observed, counted and sampled and then diluted.

2.2.2. Sample treatment
The stages of sample treatment are as follows:

a) Salmonella sp bacteria that have been obtained are then diluted with distilled water with a dilution of 106. The bacterial culture is then poured into a sterile petri dish and exposed to ultraviolet light for 15 minutes, 25 minutes and 35 minutes. Then each of these cultures was given to 0.5 ml of 1 week old ducklings orally. Furthermore, ducklings are kept in maintenance cages.

b) Two weeks after the treatment was given, each group of treatment ducks was taken for sampling. Samples taken are cloaca swabs, blood, liver and intestine. Followed by a complete hematological calculation, isolation of organs in LB media, isolation of cloacal swabs and preparation of preparations. Every two weeks surgery is performed for data collection until the eighth week.

2.2.3. Hematological examination
For hematological examination, counting white blood cell counts and, Lyposit. Blood is drawn directly from the slaughter process in the jugular vein. 3 ml of blood is taken and then put into EDTA Vaculab and stored in a coolerbox. The blood is then taken to the laboratory for examination.
Anesthetized ducks are slaughtered in the neck by severing the jugular vein. Dead ducks are then dissected to take samples of the liver and intestines. The sample is put in a container that already contains NBF with the aim of preventing damage to the cells and tissues making up the organ. Furthermore, making preparations with microtechnics. The finished preparations are then observed under a microscope with magnification 10 times or 40 times and documented.

3. Results and Discussion

3.1. Hematology White Blood Cell (WBC) and Lymphosit

Complete hematological examination functions to check health conditions such as infection or bleeding and diagnose an illness. Measured hematological conditions can be used as a basis for determining a health condition. Based on the results of the hematological examination carried out, it was seen that the number of white blood cells in the control group (without irradiation) would rise in the fourth and sixth week after the administration of bacteria and fell again in the eighth week (Table 1). This happens because the development of Salmonella bacteria in the body will increase in the fourth to sixth week. This increase in bacteria must be countered by the presence of white blood cells which have a role in protecting the body from infection and playing a role in immunity.

The number of lymphocytes will also increase with time of observation (Table 2). The presence of lymphocytes is very important in phagocytizing bacteria that enter the body. The number of bacteria will increase in the fourth and sixth week, so the body will also produce more lymphocytes to phagocyte the bacteria that are there. Leukocytes or white blood cells originate from the bone marrow and circulate throughout the bloodstream. They are an important part of our immune system. This part of the blood is capable of producing antibodies to fight foreign organisms (viruses, bacteria, and parasites) as a defense against infection, respond to allergies, and support immune function.

Table 1. White Blood Cells of Duck after being given Salmonella sp

| No | Code | WBC (x10⁵/µL) |
|----|------|---------------|
|    |      | 2 weeks after treatment | 4 weeks after treatment | 6 weeks after treatment | 8 weeks after treatment |
| 1  | K    | 228.3 | 195.1 | 185.3 | 171.2 |
| 2  | P0   | 231.5 | 161.8 | 175.6 | 177.9 |
| 3  | P1   | 204.8 | 157.7 | 166.8 | 164.4 |
| 4  | P2   | 212.4 | 120.8 | 173.1 | 115.1 |
| 5  | P3   | 164.4 | 169.4 | 171.8 | 123.3 |

Table 2. Lymphosit of Duck after being given Salmonella sp

| No | Code | Lymphosit (%) |
|----|------|---------------|
|    |      | 2 weeks after treatment | 4 weeks after treatment | 6 weeks after treatment | 8 weeks after treatment |
| 1  | K    | 58 | 57 | 71 | 84 |
| 2  | P0   | 56 | 52 | 66 | 85 |
| 3  | P1   | 74 | 33 | 50 | 84 |
| 4  | P2   | 75 | 44 | 62 | 85 |
| 5  | P3   | 64 | 34 | 22 | 82 |

3.2. Histology Overview of the Liver

Based on histological observations of duck liver eight weeks after being given the Salmonella sp bacteria found bleeding, core pycnosis, necrosis and inflammation. In all treatments there was a slight damage (Figure 1-6).
Histological conditions of the liver in all treatments showed some damage, namely bleeding, core pycnosis, necrosis and inflammation. However, it did not cause severe damage. This is because the liver has the ability to fight viral and bacterial infections that enter the body so that the Salmonella bacterial infection has not affected the liver too much as one of the digestive glands. The finding of liver damage in all treatments was caused by the bacteria Salmonella sp which was inserted into the body of the duck could invade the duck organ and affect liver function.

Bleeding occurs because of damage to the epithelial lining and lamina propria resulting in cell damage and cause blood to enter the cell. Blood flowing through intestinal capillaries from the intestine to the liver through the portal vein carries a lot of bacteria. Bacteria that flow through the blood are cleansed by Kupffer cells, which are large phagocytic macrophages when blood passes through the sinuses. Only about 1 percent of bacteria that enter the portal blood from the intestine make it through the liver into the systemic circulation [7]. Bacteria that enter are thought to cause liver damage.

Other liver damage is the occurrence of nucleus pycnosis, which is the condition of the cell nucleus shrinking and solidifying so that it is dark in color. This happens because of a core rupture and damage to chromatin, which is naturally caused by a bacterial attack. There is also a nucleus of liver cells that experience necrosis, namely cell damage or death due to infection, poison or trauma that causes irregular cell components. The condition of duck intestine after treatment is found to be some damage to the duck intestine, namely the intestinal epithelium is desquamated and goblet cell proliferation occurs. Epithelial desquamation is the process of peeling off or peeling off the epithelium so that cells will accumulate and stick to one another. Excessive desquamation can cause bleeding because it causes thinning of the epithelial lining.

Another damage that occurs in the intestinal epithelium is that goblet cells undergo proliferation. Goblet cell proliferation occurs because goblet cells release too much mucus as a reaction in the face of
the entry of bacteria into the intestine. Goblet cells that undergo proliferation appear to be larger in size than normal.

4. Conclusion

Based on the results of research that has been done, it can be concluded that the radiation of Salmonella sp using UV light for 35 minutes caused a number of histological abnormalities in the liver and intestines but was still in a condition of mild damage because only minor damage was found. Abnormalities that occur in the liver include bleeding, nucleus picnosis, necrosis and inflammation, while in the intestine found abnormalities include epithelial cell desquamation, bleeding and goblet cell proliferation.

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