Asian Journal of Aquatic Sciences, December 2023. Vol 6, Issue (3) 379-388 e-issn: 2716-4608
p-issn: 2655-366X

HAEMATOLOGY OF SALINE TILAPIA (*Oreochromis niloticus)* FED WITH MORINGA LEAF POWDER ENRICHED PELLETS

Nida Farah Afifah[1]*, Windarti[2], Herman Sarumaha[1]
[1]Department of Aquaculture, Sekolah Tinggi Perikanan dan Kelautan Matauli, Pandan 22583 Indonesia
[2]Department of Aquatic Resources Management, Faculty of Fisheries and Marine, Universitas Riau, Pekanbaru 28293 Indonesia
*nidafaraha@gmail.com

ABSTRACT

Moringa leaves (*Moringa oleifera*) contain flavonoids and saponins that can help improve fish health. Fish health and immune conditions can be predicted based on fish haematology. The study aimed to determine the effects of powder addition of Moringa leaves in fish feed pellets on the haematological disorders of saline tilapia. It was carried out from July to August 2022. Moringa leaf flour was mixed well with pelleted feed, and the dose of Moringa leaf flour was as follows: 0 (Control, without the addition of Moringa leaves), P1 (10 g/kg), P2 (15 g/kg), and P3 (20 g/kg). Fish were stored in round plastic containers (25 L, equipped with an aerator and circulation pump) for 42 days, 15 fish/container. During the study, fish were fed three times/day at satiation. Sampling was carried out twice, namely before treatment and at the end of the experiment (day 42). The results showed that adding Moringa leaf flour to fish feed pellets affected fish haematology. Fish fed with turmeric-enriched pellets showed a higher percentage of lymphocytes, increasing the fish's immunity. The best treatment was P2, which showed 81.66% lymphocytes, 7.11% monocytes, 8.66% neutrophils, and 2.55% thrombocytes. Data obtained showed that fish fed with Moringa leaf pellets had better haematological conditions than fish not fed with Moringa leaf pellets.

Keywords: Fish Blood, Lymphocytes, Immunity, Fish Supplement

1. INTRODUCTION

Tilapia (*Oreochromis niloticus*) is one of the aquaculture commodities that is quite popular with the public. This fish is considered relatively easy to cultivate and has been successfully marketed because of its many advantages. Tilapia is included in the ten priority commodities of aquaculture nationally, and the market demand for tilapia is increasing, so the development of tilapia cultivation has spread to brackish waters and the sea known as saline tilapia.

Saline tilapia has a good meat texture, is delicious and savoury, is a source of animal protein, and is quite affordable. In its habitat, saline tilapia can live in waters with a wide salinity range, so salty tilapia is also called euryhaline. Based on these advantages, saline tilapia is one of the commodities considered and developed in the aquaculture sub-sector. Aquaculture development is inseparable from various obstacles, such as slow fish growth, disease attacks, the impact of environmental pollution, and the potential mortality of cultured fish. All these types of problems often hamper the effectiveness of aquaculture and reduce the amount of production. Ineffective aquaculture and low output will impact the ability to meet the community's needs or market demand, which tends to be higher.

Farmers often make efforts to overcome fish diseases by using antibiotics. Prolonged use of antibiotics can negatively impact aquaculture activities, such as the

Received : 25 October 2023
Accepted : 10 November 2023

increase in antibiotic-resistant bacteria and pollution of the aquatic environment. In addition, the use of antibiotics in large-scale aquaculture is less efficient because antibiotics are expensive. Because of this, alternatives to antibiotics are needed as a treatment and prevention of fish diseases. One option to overcome these problems is to use natural ingredients, namely moringa leaves.

Moringa leaves have many benefits and can be used as food and medicine. Moringa leaves contain flavonoids, alkaloids, phenols, and saponins[1]. Flavonoids have a role as antioxidants and can stop free radical reactions, while saponins function as immunostimulating agents[2]. Research by Oluduro[3] has conducted moringa leaf extract can inhibit Gram-negative bacteria (*Escherichia coli* and *Salmonella typhi*) and Gram-positive (*Staphylococcus aureus* and *Enterococcus* sp) and has a role in increasing endurance that can increase the survival rate of fish so that the addition of moringa leaves to feed can be used as a supplement to increase the endurance of fish.

The increased endurance of the fish can be seen in the fish's blood. According to Amlacher *in* Rahma et al.[4], blood is one way to diagnose the health status of fish. Blood cells have an essential role in the body's defence system that can ward off disease attacks and foreign substances that enter the fish's body. Adding moringa leaves to feed is one alternative to increase fish's immune system. However, there still needs to be more data on the blood picture of saline tilapia treated with feed with moringa leaf flour, so research needs to be done to obtain this information.

2. RESEARCH METHOD

Time and Place

This research was conducted from March to May 2022 at the Marine Chemistry Laboratory, Matauli College of Fisheries and Marine Sciences.

Method

The method used is an experimental or direct observation of the research object. The design used is a completely randomized (CRD) using 1 factor, four levels of treatment, and three replications. The medicines used are P0: Control (Feeding without additional moringa flour), P1: Addition of moringa flour to feed at a dose of 10 g/kg, P2: The addition of moringa flour to the feed at a dose of 15 g/kg, and P3: The addition of moringa flour to the dinner at a dose of 20 g/kg.

Procedure

Preparation of Research Containers

The maintenance container used is a 25 L jar of 12 units. Before use, the jar is first cleaned to avoid microorganisms and pathogens. The pot is filled with water to the maximum desired limit for two days and equipped with an aerator where filling the water serves to precipitate impurities in the water.

Adaptation of Test Fish

The test fish used were tilapia seeds measuring 4-6 cm, with 180 fish obtained from Fish Farm, a cultivator in Tapanuli Tengah, before being put into the container for the adaptation stage. Tilapia seeds were first weighed using analytical scales, and body length was measured using a ruler. Tilapia seeds were put into each jar with a 15 fish/jar density. Tilapia seeds are adapted for nine days to avoid stress against salinity changes. To get 20 ppt, it is done gradually. Suppose increasing five ppt with a volume of 15 L of water, 4 L of seawater is needed. Seawater is added every two days to avoid stress on fish in raising the salinity of seawater, which must be measured using a Refractometer.

Making Moringa Leaf Flour

Moringa leaves are used in manufacturing Moringa leaf flour according to the Moringa leaf flour method[5]. Moringa leaves used in the manufacture of moringa leaf flour are green leaves picked from the

branches of trees that are approximately from the first petiole (under the shoot) to the seventh petiole that is still green. However, old leaves can be used if the moringa leaves have yet to turn yellow. The moringa leaves are washed first to remove those attached to the leaves, then the moringa leaves are dried in the sun then the moringa leaves are mashed using a blender and then sieved with a sieve to obtain moringa leaf flour and then stored in a plastic container.

Preparation of Test Feed

The ingredients of the test feed components consist of commercial pellets, moringa leaf flour, and tapioca flour as an adhesive between the shots and moringa leaf flour. The method used in making feed is the coating method, using water at an average temperature added with two spoons of tapioca flour feed and then stirred, then pouring enough boiling water while stirring until thickened. Moringa leaf flour was added and mixed in the pellets according to the treatment dose, namely P1 10 g/kg, P2 15 g/kg, and P3 20 g/kg. The feed that has been mixed is then dried in the sun until dry during the drying process. The pellets that stick to it are then separated by squeezing, and the dried pellets are then stored in the fish feed container.

Fish Maintenance

The test fish were reared for six weeks in 25 L jars equipped with aerators with a salinity of 20 ppt. During maintenance, the test fish fry was given feed that had been mixed with Moringa leaf flour. Feeding was done ad satiation with a frequency of three times a day at 08.00, 13.00, and 18.00 WIB.

Fish Blood

Blood collection of test fish was carried out twice, namely initial blood collection before treatment and final fish blood collection after treatment. The test fish used were nine fish/treatments. Fish were first anaesthetized with clove oil (about five drops/L) in a water-filled jar and stirred gently. The syringe and syringe were moistened with 10% EDTA to prevent blood clotting. Fish blood was collected through the caudal vein.

Leukocyte Differentiation

Calculation of leucocyte types based on the Blaxhall & Daisley *in* Kurniawan et al.[6] by taking fish blood, then making a blood test preparation on a glass object and then drying it, then fixing it with methanol solution for 5 minutes, after which it is rinsed with distilled water and then dried, and continued with Giemsa staining for 20 minutes, after which it is washed with running water, then dried, then observed under a microscope with a magnification of 10x40. The types of leucocytes observed were lymphocytes, monocytes, and neutrophils. Then, the leucocyte differentiation was calculated with the following formula:

$$\text{Cell percentage} = \sum n \times 100\%$$

Description:

$\sum n$ = Number of leukocyte cells

Blood cell morphology

In this study, the blood cells observed were red blood cells and various white blood Cells. For observation, blood cells prepared with Giemsa dye and stained were used. Blood cell morphology was observed using an Olympus CX 21 binocular microscope with a micrometre. The parameters observed were cell diameter size (mm), cytoplasmic and nuclear colour, and general morphological and nuclear shape[7].

Data Analysis

Leukocyte differentiation data are presented in tabular form and analyzed using analysis of variance (ANOVA), and saline tilapia blood morphology data are shown in the form of images, which are then analyzed descriptively. Survival data are presented in tabular form and analyzed using analysis of variance (ANOVA.

3. RESULT AND DISCUSSION

Blood Cell Morphology in Saline Tilapia (*O.niloticus*)

Erythrocyte cells of saline tilapia found in this study have an average diameter of 0.00942 mm or 9.42 µm and have an oval shape with an oval-shaped nucleus as well, with Giemsa staining the cytoplasm is pink. In contrast, the heart is purple/red; the Number of erythrocyte cells in each fish varies and is usually influenced by the stress and temperature of the environment. The erythrocyte cell morphology is examined to determine the morphological changes of red blood cells treated with feed using moringa leaf flour with different doses. The shape of erythrocyte cells in saline tilapia treated with moringa leaves and control showed no difference. This proves that moringa leaves do not affect the shape or structure of erythrocyte cells. The morphology of erythrocyte cells in saline tilapia from the beginning to the end of observation using a microscope has a regular body. It does not change the shape or size of saline tilapia erythrocyte cells.

The erythrocyte morphology of saline tilapia fish in this study is by the description of erythrocytes according to Mastuti[8], the morphology of mature erythrocyte cells is oval with ovoid nuclei and pink cytoplasm (Figure 1).

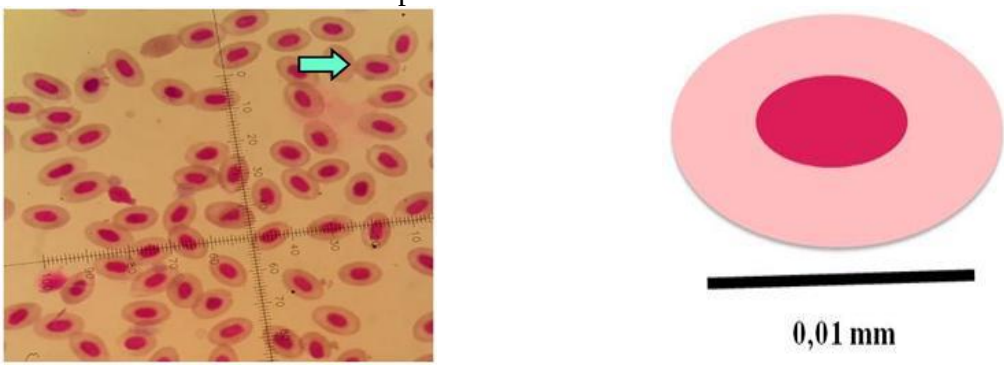

Figure 1. Erythrocyte cell morphology (1000x magnification)

Figure 2. Morphology of single and clustered lymphocyte cells (1000x magnification)

Lymphocytes

The results of observations in this study of lymphocyte cell morphology in saline tilapia show that two types of lymphocyte cells are found, namely single lymphocyte cells and clustered lymphocyte cells. Lymphocyte cells have a round or oval shape. The size of single lymphocyte cells with a diameter of 0.00700-0.00900 mm or 7-9 µm usually has a larger size than clustered lymphocyte cells, which are around 0.00400-0.00500 mm or 4-5 µm, in clustered lymphocyte cells numbering between 2-6 cells, lymphocytes have a nucleus that occupies most of the cell, so the cytoplasm does not look so clear, clustered or single lymphocytes do not contain granules with Giemsa staining blue or dark purple (Figure 2).

Monocytes

From the observations in this study, the morphology of monocyte cells in saline tilapia fish generally has a larger shape than other leukocytes. The cell nucleus is usually large and round in the middle or on the edge of bright pink, and the cytoplasm has no granules. The cytoplasm in monocytes is

irregular because it is amoeboid. With Giemsa staining, the cytoplasm of monocytes is pink to clear, and the average size of monocyte cells is 0.01036 mm or 10.36 µm.

The observation results in this study showed that the monocytes of saline tilapia fish were by the description of monocytes according to Hoffman *in* Erika[10], monocytes in fish have almost the same morphology as monocytes in mammals, oval or round, with a diameter between 8 - 15 µm. The nucleus is oval, located near the edge of the cell, and fills part of the cell contents. The monocyte nucleus is sometimes found in the centre, and the cytoplasm has no granules. The shape of monocyte cells in saline tilapia treated with moringa leaf flour in the feed showed no effect on the structure or form of monocyte cells. Figure 3 below shows monocyte cell morphology in saline tilapia obtained in this study.

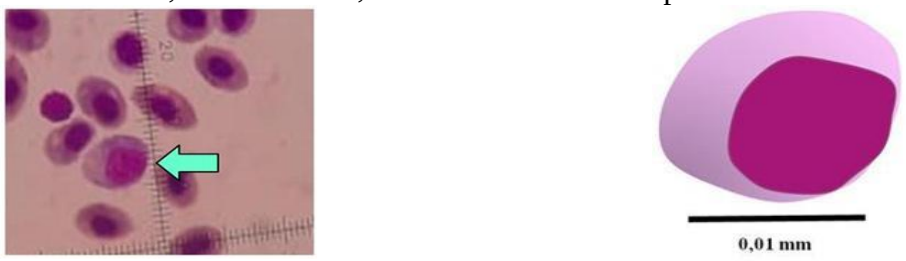

Figure 3. Monocyte cell morphology (1000x magnification)

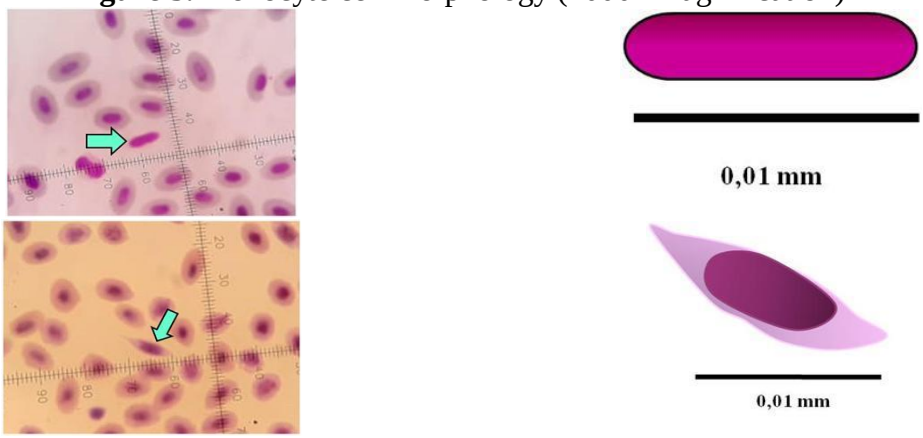

Figure 4. Platelet cell morphology (1000x magnification)

Platelets

In this study, there are two types of platelets, namely platelet cells that are oval and shaped like a cigar and platelet cells that are fusiform and shaped like a leaf. Platelet cells have clear cytoplasm, so they are not visible, and have a nucleus with purple or blue solid Giemsa staining, with an average size diameter of 0.00700-0.00900 mm or 7-9 µm (Figure 4).

The morphology of platelet cells in this study is by the description of monocytes according to Chinabut *et al. in* Dipongtonung[11] said that platelets in fish are elongated round or oval and play a role in the blood clotting process because they participate in activating prothrombin into thrombin. A unique feature of platelets is the presence of a thin cytoplasmic circle around the core, which will be dark purple when stained with Giemsa. The average size of platelets ranges from (4 x 7µm) - to (5 x 13 µm). This shows that Moringa leaf powder does not affect the structure or shape of platelet cells.

Neutrophils

The results of observations in this study of neutrophil cell morphology in saline tilapia were found to be round with a kidney-like or C-shaped nucleus with pink Giemsa staining, pale pink to colourless cytoplasm, and visible granules, having an average size of 0.01022 mm 10.22 µm. Neutrophil cell morphology in saline tilapia can be seen in Figure 5.

The basophil cells of saline tilapia in this study are the basophil cells described by Metin[12]. Basophils are smaller than heterophils and eosinophils. Basophils are dark blue granular with various sizes, and the nucleus is in the centre. Nabib and Pasaribu *in* Erika[8] also said that eosinophils and basophils are rarely seen in fish blood circulation. The shape of basophil cells in saline tilapia treated with moringa leaf flour and control showed no difference. This indicates that moringa flour does not affect the structure or form of basophil cells.

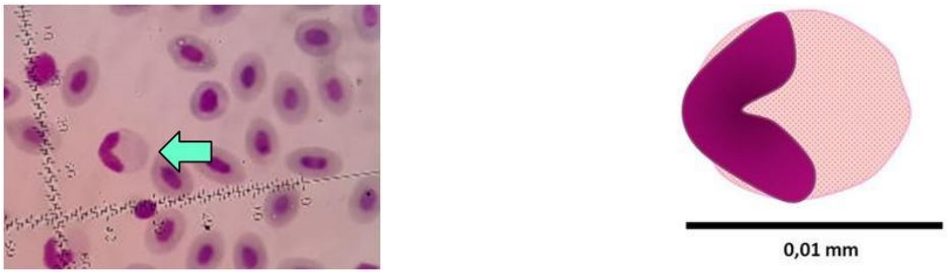

Figure 5. Neutrophil cell morphology (1000x magnification)

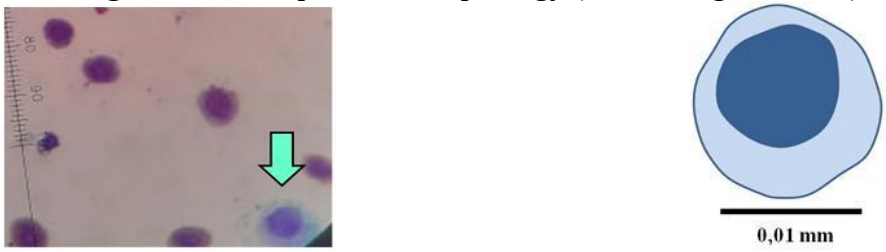

Figure 6. Basophil cell morphology (1000x magnification)

Table 1. Leukocyte differentiation in saline tilapia.

Leukocyte Differentiation	Treatment	Lymphocytes (%)	Monocytes (%)	Neutrophils (%)	Platelets (%)
Beginning of Research		70,00±0,00[a]	11,33±2,08[b]	10,00±2,00	8,66±1,52[c]
End of Research	P0	73,00±0,33[b]	10,78±0,84[b]	9,77±0,50	6,44±0,50[b]
	P1	75,88±0,50[c]	8,66±0,33[a]	9,55±0,50	5,88±1,50[b]
	P2	81,66±0,33[e]	7,11±0,50[a]	8,66±0,66	2,55±0,50[a]
	P3	78,77±0,69[d]	8,33±0,33[a]	8,66±0,33	3,44±0,69[a]

Description : Different superscript letters in the same column indicate significant differences (P<0.05) between treatments; ± Standard Deviation (SD). P0 (control), P1 (dose of moringa leaves 10 gr/kg feed), P2 (15 gr/kg), and P3 (20 gr/kg).

Observation of Leukocyte Differentiation of Tilapia

The calculation results of leukocyte differentiation were carried out to see the increase in the percentage of leukocyte types after raising saline tilapia for 42 days fed with moringa flour. The kinds of leukocyte differentiation found in this study will then be calculated as the percentage of each cell, namely five leukocyte cells: lymphocytes, monocytes, neutrophils, and platelets (Table 1).

The percentage of lymphocyte cells after 42 days of maintenance has increased, ranging from 75.88-81.66%. This shows that the addition of moringa flour can increase the production of lymphocytes in the blood. Lymphocytes function to process the mechanism of antibody formation in fish and become a defence system from the attack of foreign objects that will enter the fish body, according to the normal range of lymphocyte percentage in fish[13]. The average range of lymphocyte percentage in tilapia is 68-86%. The increase in the

Number of lymphocyte cells in white blood cells is due to the entry of compounds that act as immunostimulants[14]. According to Rosidah et al.[15], the ability of moringa flour to activate immunomodulators that modulate the components of the immune system.

The percentage of monocyte cells of saline tilapia after being fed with feed containing moringa leaf flour decreased. Monocytes that tend to fall every week are related to the function of monocytes as macrophages, where monocytes are not needed to phagocytize because no infection enters the body that stimulates monocyte production[16].

The study's results after 42 days of maintenance showed that the percentage of monocyte cells of saline tilapia changed where the highest was in the P0 treatment, which was 11.33%, and the lowest rate was in the P2 treatment, which was 7.11%. The low percentage of monocyte cells in saline tilapia proves that the fish is healthy and not attacked by destructive pathogens that enter the fish's body, so producing monocyte cells in large quantities is unnecessary. In addition, the presence of active compounds in moringa flour plays a role in suppressing bacterial infections. These active compounds are flavonoids, tannins, and saponins, thus helping the work of monocytes. Flavonoids are known to have anti-bacterial activity by carrying out mechanisms, namely inhibiting the synthesis of nucleic acids from bacteria, damaging the function of the cytoplasmic membrane, inhibiting the metabolism of bacteria, inhibiting cell membrane synthesis and aggregating bacterial cells[17].

The percentage of monocyte cells of saline tilapia after being fed with feed containing moringa leaf flour decreased. Monocytes that tend to fall every week are related to the function of monocytes as macrophages, where monocytes are not needed to phagocytize because no infection enters the body that stimulates monocyte production[16].

The study's results after 42 days of maintenance showed that the percentage of monocyte cells of saline tilapia changed where the highest was in the P0 treatment, which was 11.33%, and the lowest rate was in the P2 treatment, which was 7.11%. The low percentage of monocyte cells in saline tilapia proves that the fish is healthy and not attacked by destructive pathogens that enter the fish's body, so producing monocyte cells in large quantities is unnecessary. In addition, the presence of active compounds in moringa flour plays a role in suppressing bacterial infections. These active compounds are flavonoids, tannins, and saponins, thus helping the work of monocytes. Flavonoids are known to have anti-bacterial activity by carrying out mechanisms, namely inhibiting the synthesis of nucleic acids from bacteria, damaging the function of the cytoplasmic membrane, inhibiting the metabolism of bacteria, inhibiting cell membrane synthesis and aggregating bacterial cells[17].

The increase in the Number of neutrophil cells in fish indicates the activity of neutrophil cells in reaching and attacking antigens (foreign particles) entering the body, which suggests the phagocytosis process. The method of decreasing the percentage of neutrophil cells indicates the fish's immune system and immune system in healthy fish conditions, due to the absence of pathogenic infection attacks that enter the fish body, there is no need to phagocytose the pathogenic infection because there is no stimulus to produce neutrophil cells, this is by the opinion of Utami et al.[18], stating that an increase in the Number of neutrophils is a result of immune mechanisms that work in response to infection in the body. A low neutrophil percentage indicates no microorganism attack, so neutrophils have not been produced by the fish body[19].

Survival of Saline Tilapia

The study's results showed survival data of saline tilapia during the survey. Survival of saline tilapia in all treatments

showed the same pattern. From week 0 to week 6, the survival rate of each treatment was above 95%. The survival graph can be seen in Figure 7.

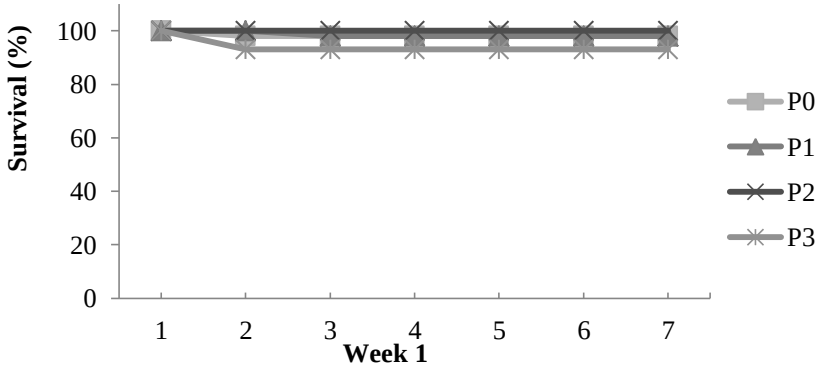

Figure 7. Survival rate of tilapia

Based on the study results in Figure 7, it can be seen that the survival of fish from all treatments is relatively high, which is above 90%. In the P2 treatment, the survival rate was 100%. In treating P0 and P1, the survival rate was 98%, and in P3, the survival rate was 93%. The high survival rate of saline tilapia fish seeds is due to favourable environmental conditions and regular feeding with adequate nutritional content. According to Iskandar & Elrifadah[20], The survival of saline tilapia is determined by environmental conditions and feed. In this study, water quality was maintained by pipetting twice daily, namely in the morning and evening, so the water remained clean and maintained. The food given is commercial pellets added with Moringa leaf flour. These commercial pellets contain 39-40% protein, and adding moringa flour will enrich the nutrition of the feed. Thus, the saline tilapia fry in this study lived in a good environment and got good food so that the fish could grow and survive well.

4. CONCLUSION

From the results obtained, feeding with the addition of moringa flour has influenced changes in the composition of leukocytes in saline tilapia. Giving moringa leaf flour to saline tilapia can increase the percentage of lymphocyte cells, which means it can boost immunity in fish so that fish are resistant to disease attacks and can reduce the rate of monocytes and neutrophils. The best dose is in the P2 treatment by giving 15 gr/kg moringa leaf flour to the feed with a lymphocyte percentage of 81.66%, monocytes 7.11%, neutrophils 8.66%, and platelets 2.55%.

REFERENCES

1. Arora, S.D., Onsare, G.J., Kaur, H. Bioprospecting of Moringa (*Moringa ceae*): Microbiological Perspective. *Journal of Pharmacognosy and Phytochemistry*, 2013; 1(6): 193
2. Bamishaiye, E.I., Olayemi, F.F., Awagu, E.F., Bamshaiye, O.M. Proximate and Phytochemical Composition of *Moringa oleifera* Leaves at Three Stages of Maturation. *Advance Journal of Food Science and Technology*, 2011; 3(4): 233-237.
3. Oluduro, A.O. Evaluation of Antimicrobial Properties and Nutritional Potentials of *Moringa oleifera* Lam. Leaf in South-Western Nigeria. *Malaysian Journal of Microbiology*, 2012; 8(2): 59-67.
4. Rahma, F.W., Mahasri, G., Surmartiwi, L. Pengaruh Pemberian Ekstrak *Sargassum* sp. dengan Pelarut Metanol pada Pakan terhadap Jumlah Eritrosit dan Differensial Leukosit

Ikan Lele Dumbo (*Clarias gariepinus*). *Jurnal Ilmiah Perikanan dan Kelautan*, 2015; 7(2): 213-218.

5. Zakaria., Tamrin, A., Sirajuddin., Hartono, R. Penambahan Tepung Daun Kelor pada Menu Makanan Sehari-Hari dalam Upaya Penanggulangan Gizi Kurang pada Anak Balita. *Media Gizi Pangan,* 2012; 13(1): 41-47.
6. Kurniawan, R., Syawal, H., Effendi, I. Pengaruh Penambahan Suplemen Herbal pada Pakan terhadap Diferensiasi Leukosit Ikan dan Sintasan Ikan Patin (*Pangasionodon hypopthalmus*). *Jurnal Akuakultur Rawa Indonesia*, 2020; 8(2): 150-163.
7. Windarti. *Keterampilan Dasar Biologi Perikanan*. Fakultas Perikanan dan Kelautan Universitas Riau. Riau, 2020; 160pp.
8. Mastuti, R. *Pengobatan Penyakit MAS (Motile Aeromonas Septicaemia) dengan Ekstrak Daun Mangrove (Rhizophora sp.) pada Ikan Jambal Siam (Pangasius hypophthalmus).* Pekanbaru: Fakultas Perikanan dan Kelautan Universitas Riau, 2017.
9. Hernawati, R.D. Study of The Effects of Carboxymethyl Chitosan on The Non-specific Defense System in the Carp (*Cyprinus carpio*). *Jurnal Sain Veteriner,* 2013; 31(1).
10. Erika, Y. *Gambaran Diferensiasi Leukosit pada Ikan Mujair (Oreochromis mossambicus) di Daerah Ciampea Bogor.* Fakultas Kedokteran Hewan, Institut Pertanian Bogor, 2008.
11. Dopongtonung, A. *Gambaran Darah Ikan Lele (Clarias spp.) yang Berasal dari Daerah Laladon Bogor.* Fakultas Perikanan dan Ilmu Kelautan. Institut Pertanian Bogor. 2008.
12. Metin, K.Y., Basimoglu, K,F., Kargin, K.S., Koca, O., Turkozan. Blood Cell Morphology and Plasma Biochemistry of Captive *Mauremys caspica* (Gmelin, 1774) and *Mauremys rivulata* (Valenciennes, 1833). *ActaVet. Brno*, 2008; 77: 163-174.
13. Hardi, E.H., Sukenda, Harris, E., Lusiastuti, A.M. Karakteristik dan Patogenitas *Streptococcus agalactiae* tipe ß- hemolitik dan Non-Hemolitik pada Ikan Nila. *Jurnal Veteriner*, 2011; 12(2): 152-164.
14. Rustikawati, I. *Efektifitas Ekstrak Sargassum sp. dalam Meningkatkan Imunitas Ikan Nila (Oreochromis niloticus) terhadap Serangan Streptocciasis.* Program Pasca sarjana Universitas Padjadjaran Bandung, Bandung, 2011: 84pp.
15. Rosidah, I.D., Buwono, W., Lili, I.B., Suryadi, S., Reza, A. Ketahanan Ikan Lele Sangkuriang, *Clarias gariepinus* (Burchell, 1822) terhadap *Aeromonas hydrophila* Pasca Pemberian Ekstrak Daun Kelor (*Moringa oleifera* L) melalui Pakan. *Jurnal Iktiologi Indonesia*, 2018; 19(1): 97–113.
16. Ginting, K.D., Riauwaty, M., Syawal, H. Diferensiasi Leukosit Ikan Lele Dumbo (*Clarias gariepinus*) yang diberi Pakan Mengandung Kunyit *(Curcuma domestica* Val.) dan Diinfeksi Bakteri *Aeromonas hydrophila*. *Jurnal Ilmu Perairan (Aquatic Science),* 2021; *9*(2), 116-125.
17. Kenconojati, H., Rukmana, N.R. Daya Hambat Ekstrak Daun Kelor (*Moringa oleifera*) terhadap *Aeromonas hydrophila*: Studi Awal untuk Pengobatan *Aeromoniasis*. *Journal of Aquaculture Science*, 2019; 4(1): 12-20.
18. Utami, D.T., Prayitno, S.B., Hastuti, S., Santika, A., Gambaran parameter hematologis pada ikan nila (*Oreochromis niloticus)* yang diberi vaksin DNA *Streptococcus iniae* dengan dosis yang berbeda. *Journal of Aquaculture Management and Technology*, 2013; 2(4): 7-20
19. Hartika, R., Mustahal, M., Putra, A.N. Gambaran Darah Ikan Nila (*Oreochromis niloticus*) dengan Penambahan Dosis Prebiotik yang Berbeda dalam Pakan. *Jurnal Perikanan dan Kelautan,* 2014; 4(4): 259-267.

20. Iskandar, R., Elrifadah, E. Pertumbuhan dan Efisiensi Pakan IkanNila (*Oreochromis niloticus*) yang Diberi Pakan Buatan Berbasis Kiambang. *Jurnal Ziraa'ah*, 2015; 40(1): 18–24