The use of kailan \((Brassica oleracea\ L.)\), lettuce \((Lactuca sativa\ L.)\) and pakcoy \((Brassica rapa\ L.)\) in the cultivation of striped catfish \((Pangasianodon hypophthalmus)\) aquaponic system on blood glucose levels and oxygen consumption levels

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Abstract. In striped catfish cultivation water quality greatly affects the condition of fish. The use of an aquaponic system can improve the water quality, because the basic principle of aquaponics is to utilize fish waste and fish feed residues that are not consumed and it can be used as a nutrients for kailan, lettuce and pakcoy. The purpose of this study was to determine whether the three plants can affect blood glucose levels and the level of oxygen consumption of catfish. The research method using an experimental method with a Completely Randomized Design (CRD) consisting of 4 treatments and 5 replications. At P0 are control (without plants), P1 (kailan), P2 (lettuce), and P3 (pakcoy). The main parameters observed were blood glucose levels and oxygen consumption levels. While the supporting parameters are measurement of temperature, DO, pH, plant measurements at the beginning and end of the study. The results showed that there was a significant difference \((p<0.05)\) on blood glucose levels and oxygen consumption levels in aquaponic system cultivation. The treatment that gave the best value for blood glucose levels and oxygen consumption was the use of lettuce plants (P2).

1. Introduction
Striped catfish \((Pangasianodon hypophthalmus)\) is one of the consumption freshwater fishery products that are of interest to the Indonesian people and has important economic value in the world of aquaculture. The Department of Fisheries and Aquaculture FAO (Food and Agriculture Organization) states that catfish ranks fifth after carp \((Cyprinus carpio)\), tilapia \((Oreochromis niloticus)\), catfish \((Clarias sp.)\) and gourami \((Osphronemus gouramy)\) \([1]\). Based on data from the \([2]\) striped catfish production in 2015 was 339,069 tons, in 2016 it was 392,918 tons, in 2017 it was 319,966 tons and in 2018 it was 391,151. From these data, it is known that the production of striped catfish from 2015 to 2018 has increased by an average of 6.52%.

Increased production of striped catfish indicates that striped catfish have good prospects for cultivation because striped catfish have the advantages of being easy to maintain, the taste of the meat is delicious and savory, the individual size of catfish is quite large, growth is relatively fast, and has high economic value \([1]\). With the high demand for catfish cultivation, it causes obstacles for urban areas, namely limited land and shrinking water sources. So to overcome this, you can take advantage of the use of an aquaponics system.
According to Kyaw and Andrew [3], Aquaponics is a combination of fish cultivation (aquaculture) and vegetable crop cultivation without using soil (hydroponics) in one place. In principle, aquaponics combines aquaculture as the main sector of agribusiness, while crop yields are a by-product. In catfish cultivation, water quality greatly affects the condition of fish, fish can become stressed and even die due to suboptimal water quality.

One indicator of stress levels in fish can be seen from the value of blood glucose levels. Stressed fish can cause high blood glucose. According to [4], stressed fish will experience an increase in glucocorticoids which results in increased blood glucose levels due to high energy requirements during stress. In fact, fish farming activities always occur competition in the use of space and the utilization of available oxygen.

Each level of biota has differences in the level of oxygen consumption, because oxygen uptake depends on the intensity of metabolism which is influenced by body weight expressed in units of mg/g/hour. The level of fish oxygen consumption is the amount of oxygen needed by fish. Oxygen is needed to help metabolic processes in the body [5]. According to [6], increased oxygen consumption can result in reduced availability of dissolved oxygen in the environment, so oxygen needs are not met which can make fish stressed. Therefore, according to [7], the use of an aquaponic system can improve water quality, because the basic principle of aquaponics is to utilize fish waste and fish feed residues that are not consumed as nutrients for cultivated plants.

In this study, we used kailan (Brassica oleracea L.), lettuce (Lactuca sativa L.) and pakcoy (Brassica rapa L.). The use of these three plants is because they are easy to cultivate, have fibrous roots and have economic value that can be harvested and consumed. Kailan plant was chosen because it has a fairly tough resistance with a relatively short harvest time [8]. Meanwhile, according to [9], kailan plants are harvested by picking leaves so that they require a lot of nutrients for the leaf formation process. The selection of pakcoy plants is as a plant that is effective in absorbing organic matter contained in catfish culture waste. These plants are also referred to as heavy feeders because they require more nitrogen for growth [10]. And the selection of lettuce plants that have a tap and fibrous root system so that it is suitable for cultivation in an aquaponic system because the maintenance period requires water that flows continuously [11]. Meanwhile, according to [12], stated that the width of the leaves on lettuce plants can support the implementation of the photosynthesis process because there is chlorophyll. With the presence of chlorophyll, plants can independently make their own food so that they play an important role in the process of plant growth and formation.

Based on this, this study was conducted to determine the use of kailan (Brassica oleracea L.), lettuce (Lactuca sativa L.) and pakcoy (Brassica rapa L.) in aquaponic catfish (Pangasianodon hypophthalmus) cultivation on blood glucose levels and oxygen consumption rate.

2. Materials and Methods
2.1. Experimental Design
The research method using an experimental method with a Completely Randomized Design (CRD) consisting of 4 treatments and 5 replications. At P0 = control (without plants), P1 = kailan, P2 = lettuce, and P3 = pakcoy. The main parameters observed were blood glucose levels and oxygen consumption levels. While the supporting parameters are measurement of temperature, DO, pH, plant measurements at the beginning and end of the study.

2.2. Preparation of Tools and Materials
Preparation of tools and materials is the first step of a research. The equipment to be used is washed with soap and then rinsed with clean water. After that, all equipment is immersed in a chlorine solution for 24 hours which aims to make the tools to be used free from bacteria. After that, rinse with clean water until the chlorine smell is gone. The plants used in this study were kailan, lettuce and pakcoy which were 14 days old. The three plants were planted in a netpot container with rockwool as the planting medium. In each gutter there are 5 plants with a spacing of 10 cm.
2.3. Making Aquaponics System
Preparation pipes and gutters as a series of aquaponic system tools. The pipe is used to drain water from the aquarium to the plant media gutters, while the gutters function as a media for kailan, lettuce and pakcoy plants which have been equipped with inlet and outlet holes measuring 3 cm. The required gutters are 20 pieces with a size of approximately 60 cm x 15 cm x 15 cm. In each netpot hole, the plants in the gutter are 6 cm apart. The gutters and pipes are arranged above an aquarium measuring 40 x 30 x 30 cm which has been equipped with inlet and outlet channels. This is so that the waste from the aquarium will circulate through the filter and then flow into the gutters of the plant media and then back to the aquarium.

2.4. Preparation and Maintenance of Striped catfish
In this study, 360 catfish seeds were used with a size of 6-7 cm. The preparations carried out were that the fish were first acclimatized in a tank profile that was filled with air. Acclimatization is one of the methods used so that fish can adjust to their new environment, because fish will feel the difference in temperature, pH, and the amount of oxygen they get. The acclimatization was carried out for 30 minutes until the catfish fry slowly came out of the plastic office. After acclimatization, each purchase was filled with 18 fish. The maintenance of catfish fry was carried out for 28 days. Striped catfish are reared at a density of 18 fish/18 liters. During the maintenance of catfish are fed commercial pellets 2x a day, namely at 08.00 and 15.00. The amount of feed given is 3% of the body weight of the fish.

2.5. Blood Glucose Levels
Blood glucose levels in fish serve as the main energy source and an important substrate for cell metabolism, especially in the brain cells. To keep brain function working optimally, glucose is needed continuously. If the fish's blood glucose level is below or above the optimal value, then the fish is in a state of stress which can cause hyperglycemia, namely increased blood glucose levels, because fish have normal limits. According to [13], stated that normal fish blood glucose levels ranged from 40-90 mg/dL. Measurement of blood glucose levels using the GES GD-013 Glucometer. Fish blood samples are dropped on the end of the test strip and the results will appear on the screen in mg/dL units.

2.6. Oxygen Consumption Levels
Oxygen consumption rate was used to determine growth-related metabolic rate. The calculation of the level of oxygen consumption is calculated based on the [14]:

\[
\text{Oxygen consumption levels (mgO}_2/\text{g/hour)} = \frac{V(DO_0 - DO_t)}{W \times T}
\]

Information:
\[V\] = volume of air in the container (L)
\[DO_0\] = dissolved oxygen concentration at the beginning of the observation (mg/L)
\[DO_t\] = dissolved oxygen concentration at the end of observation (mg/L)
\[W\] = weight of test fish (g)
\[T\] = observation time (hours)

2.7. Water Quality Analysis
Water quality greatly affects the survival of fish, because water is the main medium for fish to live. According to [15] stated that temperature is one of the most important and influential physical factors for fish growth. A good water temperature for the growth of catfish is 28-30°C and the pH value is between 6.5 to 9. While the optimal value of water quality according to [16], namely the water temperature ranges from 26-31°C, pH 6-9, dissolved oxygen or DO >3 mg/L, and levels of ammonia (NH₃) 0.1 mg/L.
2.8. Statistic Analysis

The data obtained were analyzed using the Anova test (Analysis of Variance) to determine the effect of the treatment given. If there are significant results, the calculation is continued with Duncan's Multiple Range Test with a 5% significance level.

3. Result and Discussion

3.1. Blood Glucose Levels

The results of measuring blood glucose levels in catfish for 28 days can be seen in Table 1.

| Table 1. Average Blood Glucose Levels |
|--------------------------------------|
| Treatments   | Day7     | Day14    | Day21     | Day28     |
| P0           | 84.20b±0.837 | 86.00d±2.550 | 89.60d±2.966 | 93.40d±3.912 |
| P1           | 78.20b±0.837 | 78.80b±0.837 | 84.20b±3.962 | 88.20b±2.490 |
| P2           | 76.00a±1.581 | 75.80a±2.588 | 75.40a±1.673 | 77.40a±5.320 |
| P3           | 81.20c±1.304 | 81.60c±1.140 | 80.00b±2.739 | 82.40b±1.949 |

Note: different superscripts on the same line show significantly different effects (p<0.05)

Based on the results of the statistical test Analysis of Variance, blood glucose levels of catfish on day 7 to day 28 showed a significant difference (p<0.05) in each treatment with different types of plants. On the 7th day, the highest blood glucose level was at P0 with a value of 84.20±0.837 mg/dL and the lowest blood glucose level was at P2 with a value of 76.00±1.581 mg/dL. On day 14, the highest blood glucose level was at P0 with a value of 86.00±2.550 mg/dL and the lowest blood glucose level was at P2 with a value of 75.80±2.588 mg/dL. On day 21, the highest blood glucose level was at P0 with a value of 89.60±2.966 mg/dL and the lowest blood glucose level was at P2 with a value of 75.40±1.673 mg/dL. On day 28, the highest blood glucose level was at P0 with a value of 93.40±3.912 mg/dL and the lowest blood glucose level was at P2 with a value of 77.40±5.320 mg/dL.

3.2. Oxygen Consumption Levels

The results of measuring the level of oxygen consumption in catfish for 28 days can be seen in Table 2.

| Table 2. Average Oxygen Consumption Levels |
|---------------------------------------------|
| Treatments   | Day7     | Day14    | Day21     | Day28     |
| P0           | 0.1548a±0.0112 | 0.1712a±0.0372 | 0.2027a±0.0137 | 0.2401a±0.0167 |
| P1           | 0.0932c±0.0165 | 0.1026c±0.0071 | 0.1605b±0.0087 | 0.2014b±0.0097 |
| P2           | 0.0708d±0.0075 | 0.0665d±0.0053 | 0.0611d±0.0122 | 0.0883d±0.0031 |
| P3           | 0.1258b±0.0199 | 0.1307b±0.0054 | 0.1134c±0.0154 | 0.1370c±0.0158 |

Note: different superscripts on the same line show significantly different effects (p<0.05)

Based on the results of the statistical test ANOVA (Analysis of Variance), the level of oxygen consumption of catfish on day 7 to day 28 showed a significant difference (p<0.05) in each treatment with different plant species. On the 7th day the highest level of oxygen consumption was at P0 with a value of 0.1548±0.0112 mgO₂/g/hour and the lowest level of oxygen consumption was at P2 with a value of 0.0708±0.0075 mgO₂/g/hour. On the 14th day the highest level of oxygen consumption was at P0 with a value of 0.1712±0.0372 mgO₂/g/hour and the lowest level of oxygen consumption was at P2
with a value of 0.0665±0.0053 mgO₂/g/hour. On day 21 the highest oxygen consumption level was at P0 with a value of 0.2027±0.0137 mgO₂/g/hour and the lowest oxygen consumption level was at P2 with a value of 0.0611±0.0122 mgO₂/g/hour. On day 28, the highest oxygen consumption level was at P0 with a value of 0.2401±0.0167 mgO₂/g/hour and the lowest oxygen consumption level was at P2 with a value of 0.0883±0.0031 mgO₂/g/hour.

3.3. Water Quality
Water quality is a supporting parameter in the maintenance of catfish for 28 days. The observed water quality parameters include temperature, dissolved oxygen, pH, ammonia which can be seen in Table 3. Table 3. Range of Water Quality

| Treatments | Temperature (℃) | DO (mg/L) | pH     | Amonia (mg/L) |
|------------|-----------------|-----------|--------|---------------|
| P0         | 28 – 30.8       | 3.57 – 7.05 | 6.8 – 8 | 0.13 – 1.12   |
| P1         | 26.3 – 28.7     | 3.61 – 7.33 | 7 – 8.5 | 0.02 – 1.07   |
| P2         | 27.4 – 30.1     | 3.55 – 7.95 | 6.6 – 8.4 | 0.01 – 0.91 |
| P3         | 27.3 – 29.8     | 3.65 – 6.65 | 6.8 – 8.4 | 0.02 – 0.91   |

(SNI: 2016) 26-31 >3 6-9 < 0.1

Based on the results of the measurement of the ranged temperature in the maintenance of catfish with aquaponics system, namely in the control treatment (P0) of 28 – 30.8℃. in the treatment with kailan plants (P1) by 26.3 – 28.7℃. in the treatment with lettuce (P2) by 27.4 – 30.1℃ and in the treatment with pakcoy plants (P3) of 27.3 – 29.8℃. The results of the observation of the ranged DO in catfish rearing with aquaponics system. namely in the control treatment (P0) of 3.57 – 7.05 mg/L. in the treatment with kailan plants (P1) at 43.61 – 7.33 mg/L. in the treatment with lettuce (P2) was 3.55 – 7.95 mg/L and the treatment with pakcoy (P3) was 3.65 – 6.65 mg/L. The results of observations of the ranged pH in the maintenance of catfish with aquaponics system. namely the control treatment (P0) of 6.8 – 8. the treatment with kailan plants (P1) of 7 – 8.5. in the treatment with lettuce plants (P2) of 6.6 – 8.4 and in the treatment with pakcoy plants (P3) of 6.8 – 8.4. While the results of the ranged observation of ammonia in catfish rearing with aquaponics systems. namely in the control treatment (P0) of 0.13 – 1.12 mg/L in the treatment with kailan plants (P1) by 0.02 – 1.07 mg/L. in the treatment with lettuce. (P2) of 0.01 – 0.91 mg/L and in the treatment with pakcoy plants (P3) of 0.02 – 0.91 mg/L.

3.4. Plants Growth
The average results of measuring the growth of kailan. lettuce and pakcoy plants during the study can be seen in Figure 1.

Figure 1. Plant Growth of Kailan. Lettuce and Pakcoy

Based on the results of measurements of the growth of kailan, lettuce and pakcoy during the research period. it was found that in the P1 treatment (kailan plants) experienced growth with an average plant
height of 3.60 cm and the average number of leaves per plant increased by 1 strand. In P2 (Lettuce) the average plant height increased by 9.65 cm with an average number of 4 leaves. While in the P3 treatment (pakcoy plant) the average plant height increased by 5.42 cm with an average number of leaves of 2 leaves per plant.

3.5. Discussion

Based on the results of statistical analysis of blood glucose levels for 28 days. on the 7th day the highest blood glucose levels were at P0 with a value of 84.2 mg/dL and the lowest blood glucose levels were at P2 (lettuce plants) with a value of 76 mg/dL. This is in accordance with the statement of [13], which states that normal fish blood glucose levels range from 40-90 mg/dL. On the 14th day there was a decrease in blood glucose levels of catfish in P2 with the value of blood glucose levels being 75.8 mg/dL. Decreased blood glucose levels indicate that fish are able to utilize energy in glucose to respond and adapt to stress [17]. On the 21st day, the blood glucose levels of P0 and P1 increased from day 7 to 89.6 and 84.2 mg/dL, respectively. It is known that fish experience stress due to the buildup of leftover feed and feces which causes water quality to be not optimal [3]. Water quality is not optimal as a result of aquaculture waste such as feces and unconsumed feed residue settling on the bottom of the water so that the ammonia content increases. The increased ammonia content can cause a decrease in appetite, so the growth of catfish becomes inhibited and can cause disruption of the fish's body metabolism until death occurs [18]. On day 28, all treatments experienced an increase in the value of their blood glucose levels. The highest glucose level value was found in the control treatment or P0 which was 93.4 mg/dL, the increase was thought to be due to the stress response arising from the high ammonia level. While the lowest value of glucose levels is P2 of 77.4 mg/dL which indicates that the best value of blood glucose is P2 using lettuce plants which tend to decrease from day 7 to day 21.

The level of oxygen consumption is the amount of oxygen needed by a living organism. Oxygen is needed to help metabolic processes in the body. Therefore, the survival of fish is largely determined by the ability to obtain sufficient O₂ from the environment [5]. Based on the results of statistical analysis of consumption levels for 28 days, it showed that the level of oxygen consumption value per gram of catfish in the size range of 1.9 – 4.02 g/head ranged from 0.047 – 0.255 mg O₂/g/hour (Table 2). This is stated by [17], that the level of oxygen consumption is influenced by the size of the fish. larger fish consume more oxygen than smaller fish. This is in line with the increase in oxygen consumption starting on the 7th, 14th, 21st, and 28th days during the study period. In addition, the higher ammonia content in the water affects the oxygen consumption level of catfish. According to [19], stated that this can happen when ammonia in the waters increases, it can cause the absorption of ammonia from the environment to be disturbed, which in turn causes an increase in ammonia levels in the blood so that oxygen consumption in fish will also increase until they die.

Fish tend to move more actively to get maximum oxygen in low dissolved oxygen conditions. According to [20], stated that oxygen consumption increases if there is a more active movement of activity so that the respiration process of fish is higher. In accordance with the statement of [6], namely an increase in oxygen consumption can result in the availability of dissolved oxygen in the environment decreasing. So oxygen needs are not met which can make fish stressed. Several biotic factors such as activity, body weight, age, temperature, stressors and fasting can also affect the metabolic rate which will then affect the rate of oxygen consumption.

Water quality greatly affects the survival of fish, so the water used as a culture medium must have an optimal value in accordance with the needs of fish life. Water quality parameters that must be considered for fish farming include temperature, DO, pH and ammonia [21]. Based on the results of measurements for 28 days, the temperature ranged from 26.3°C - 30.8°C according to [16], stating that temperature is one of the physical factors of waters that is very important and affects the growth and appetite of fish. A good water temperature for the growth of catfish is around 26-31°C. This shows that the temperature in the waters is in optimal conditions.

The results of measuring the degree of acidity (pH) during maintenance ranged from 6.6 to 8.5. This is good for the life of catfish because it is in accordance with [16], which states that the optimal pH is
between 6-9. If the pH is below the optimal value or low, the dissolved oxygen content will decrease. as a result oxygen consumption will decrease. respiratory activity increases and appetite decreases [15].

In DO measurement, the results ranged from 3.55 to 7.95 mg/L which was categorized as still in normal condition. This is reinforced by [16], that dissolved oxygen or DO is good for the survival of catfish. If the pH is below the optimal value or low, the dissolved oxygen content will decrease. DO measurement, the results ranged from 3.55 to 7.95 mg/L which was categorized as still in normal condition. This is reinforced by [16], that dissolved oxygen or DO is good for the survival of catfish. Therefore, the aquaponics system affects the blood glucose levels and oxygen consumption level of the Striped catfish (Pangasianodon hypophthalmus).

According to [16], states that the optimal ammonia level is <0.1 mg/L because the ammonia content in waters that exceeds the optimal value can be toxic so that it can endanger fish life. However, based on the measurement of ammonia in this study, the results ranged from 0.01 mg/L – 1.12 mg/L. However, the ammonia level is still in a concentration that can be tolerated by catfish. This was stated by [22], who revealed that levels of ammonia that can cause toxicity ranged from 0.4 mg/L – 3.1 mg/L.

Measurement of growth of kailan, lettuce and pakcoy for 28 days was carried out at the beginning and end of the study. Based on the measurement results, it was found that the treatments P1, P2 and P3 experienced growth with an average plant height of 3.60 cm, 9.65 cm and 5.42 cm. With the average number of leaves per plant increased by 1 strand, 4 strands and 2 strands. This shows that the three plants have different absorption power. The best growth was found in lettuce (P2) because it had the most number of leaves and wider leaf width. According to [12], states that leaf width can support the photosynthesis process because there is chlorophyll. With the presence of chlorophyll, plants can independently make their own food so that they play an important role in the process of plant growth and formation. Therefore, the more the number of leaves, the more space for photosynthesis and maximum results.

4. Conclusion
The use of kailan (Brassica oleracea L.), lettuce (Lactuca sativa L.) and pakcoy (Brassica rapa L.) in the aquaponics system affects the blood glucose levels and oxygen consumption level of the Striped catfish. The value of the lowest blood glucose levels and oxygen consumption level in the use of lettuce plants (P2).

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