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Toxicity of Industrial Palm Oil Waste Water and Sub-Lethal Test to *Oreochromis* sp.

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Abstract: ***Research of Toxicity and sub-lethal test of industrial palm oil waste water to Oreochromis sp. has been conducted on February, 1st to March, 1st, 2019 in Aquaculture Technology Laboratory, Fisheries and Marine Science Faculty University of Riau. Objective of this research is to determine the concentration of a test material or the level of an agent that produces a deleterious effect on Oreochromis sp. during a short-time exposure under controlled condition. In this study, static 96-h acute toxicity tests were carried out using Oreochromis sp. as test organisms. Probit analysis using the computer software EPA and graphical method were used to calculate the 96-h LC_{50} depending on data suitability. Results of the 96-h LC_{50} and Biological Safety Level of the industrial palm oil waste water were 126, 06 ml/L and 1.26 ml/L respectively.***

Keywords: Toxicity, industrial palm oil waste water, sub lethal, *Oreochromis*

1. Background

Oil palm plantations create a variety of impacts on the surrounding environment. Impacts of oil palm include loss of native vegetation, erosion of soil, sedimentation of streams, rivers and estuaries, introduction of pesticides and fertilizer and water and air pollutants (Keu, 2000). Environmental organizations have identified the lack of independent monitoring of the oil palm industry as a major flaw in its development throughout PNG (La Franchi, 1999). An independent assessment of oil palm plantations and mills would provide important information in assessing their environmental impacts.

Industrial waste can cause the happening of pollution. Water pollution can cause damages to the environment. Expected no control over the wastewater treatment system. Basically, pollution consists of four types that are pollution of soil, water, air, food and medicines (Sastrawijaya, 2000). The process of pollution, especially water pollution can be not separated from the presence of industry. Among these are the palm oil industry is very fast development. Indonesia is state producer biggest palm oil in the world and is generally derived from Riau Province. Without have control of the industry, it is clear that the industry will waste a lot of waste in the environment.

Palm oil waste water as a pollutants potential to pollute the environment, because the waste water is odorless, contains the value of COD and BOD and suspended solids are high. For comparison that one ton of palm oil will produce two and a half tons of waste. Control necessary physical treatment, chemical and biological (Said, 1996), because there is no one piece of equipment that can be directly to detect whether a polluted waters, but can only be measured by using organism (Syafriadiman, 2000). One of the cultured organisme that canbe used as a bio-indicator organism are red tilapia (*Oreocromis*sp).

Red tilapia fish species classified as resistant to changing environmental conditions, and the fish is a fish consumption in great demand by the people of Riau and much cultivated by the owners of oil palm plantations, especially oilfields in the transhes disposal plant. Ordinary tilapia farmed in rivers, but fish in a polluted environment can cause death. As reported in the Indonesian media on Friday 8 November 2002 issue of "Thousands of Fish Dead in Pekanbaru Siak River". This pollution occurs due to the disposal of industrial waste palm oil into the Siak River. Quantity of waste generated is 120m3/day. Total volume of waste from each palm oil with a capacity of30 tons of fresh fruit bunches/day is 600m^3/d (Said, 1996).

The potential of waste palm oil has caused considerable environmental pollution. For that we need to know the effect of palm oil waste is the life of red tilapia. Furthermore, the need to find safety biological level to life tilapia waste. And is very important information to cultivate tilapia in industrial areas.

2. Research Methods

Based on the results of preliminary tests it was found that the concentration range obtained was 125-128 ml/L for the definitive test for determining test 96 hour LC50 effluent oil palm industry. The container used is the tank cleaned and washed with 20 ppm potassium permanganate (Afrianto and Liviawati 1992), and marked (labeled) as a treatment to be used. Water solvent waste solution used water taken from wells drilled Faculty of Fisheries and Marine Sciences, University of Riau and aerated for 48 hours until the dissolved oxygen concentration of about 6 ppm. Liquid waste palm oil as a toxicant derived from PTPN V VFD Oil Permai KM 55 Dayun Siak District. Solution volume for each treatment was 20 liters / aquarium. A test organism in acute and chronic toxicity test in this study is a red tilapia (*Oreochromis* sp.) purchased at Artificial Lake Hatchery Limbungan Rumbai. The average age of the seed 1 month with the size of ± 5 cm. The number of fish per aquarium is 10 cows. Before the acute toxicity test first fish acclimatized for 2 days.

Static methods used in the acute toxicity test in determining the death of 50% of the test organisms. The characteristics of dead fish characterized by the absence of tilapia movement, loss of balance and the change in the levels of growth (Rand and Petrocelli, 1985). Observations are made every single time in the last 12hours by observing the state and behavior of fish visually. Then record all the symptoms that occur in fish as well as counting the number of fish dying. 96-hour LC50 values of palm oil waste water to the Red Tilapia fish (*Oreochromis* sp.) were determined using the EPA Probit Analysis Programversion1.5. Boundary Biologycal Safety Level (BSL) was calculated using the formula proposed by Denton *et al* (*in* Syafriadiman 2000) as follows:

BSL = LC_{50}96 hx "application factor"

BSL= Biologycal Safety Level

Numbers of application factor is 0.01

Measuring the value of water quality parameters during the test growth are DO, pH, temperature, ammonia and turbidity, which is done once a week for a month. Determination of water quality parameters was conducted before fish aquarium into a container. Measuring of absolute weight and growth rate are:

1. Absolute weight (Aw), Aw=Wt-Wo (Rickers, 1975);

Where: Aw=Absolute weight (g)
Wt=Weight of fish end of the observation (g)
Wo =Weight of fish early observations (g)

2. Daily Growth Rate (α)

$$\alpha = \sqrt[t]{\frac{Wt}{Wo}} - 1 \text{ x } 100\ \% \quad \text{(Rickers, 1975)}$$

where: α =daily growth rate (%)

Wo =Weight of fish early observations (g)
Wt =Weight of fish end of the observation (g)
t =Duration Research (days)

3. Results and Discussion

The results of acute toxicity tests

Mortality seed Red Tilapia (*Oreochromis* sp.) during the acute toxicity test increased with increasing concentrations of the palm oil industrywastefor96 hours of exposure (Table 1).

Table 1: Mortality seeds of red tilapia (*Oreochromis* sp.) during the acute toxicity test

Treatment	Mortality of seeds of red tilapia (%)				
	Replications			Total	Mean
	1	2	3		
Control	0	0	0	0	0
125, 0	10	0	10	20	6, 67
125, 4	20	20	10	50	16, 67
125, 8	20	30	20	70	23, 33
126, 2	50	40	40	130	43, 33
126, 6	70	80	80	230	76, 67
127, 0	100	100	100	300	100

Table 3 shows the mortality of red tilapia seed in each treatment during the study. Mortality was 0% in controls. Percentage mortality found in this study together with the results of Romi (2003) and Jumedi (2003). Rand and Petrocelli (1985) revealed that the organism should be 0% mortality in the control treatment. The sequence of tilapia seed mortality during the 96 hours of research the toxicity of wastewater from the palm oil industry high to low concentration was 127.0 ml/L, 126.6 ml/L, 126.2 ml/L, 125.8 ml/L 125.4 ml/L and 125.0 ml/L (Figure 1). In descriptive Figure 1 shows that the lowest mortality 0% and 100% is the highest concentration of 127.0 ml/L.

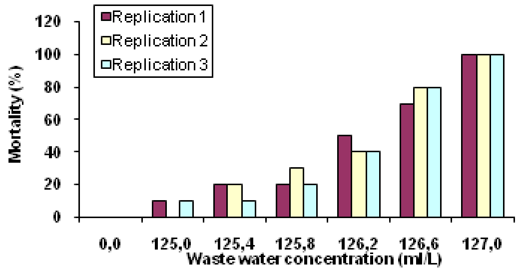

Figure 1: Histogram of seed mortality of red tilapia (Oreochromis sp) during the acute toxicity test

Analysis of variants (anova) showed that the palm oil industry waste treatment was highly significant for tilapia seed mortality ($p<0.01$). The results of Newman-Keuls test further showed that concentration of127.0ml/L very significantly ($p <0.01$) with other treatments. While the concentration of126.2ml/L did not significantly ($p>0.05$) with another treatment. Treatment126.6ml/L very significantly ($p <0.01$) with controls, significantly ($p <0.05$) with a concentration of126.2ml/L and others did not significantly ($p>0.05$). Concentrations of 126.2ml/L, 125.8ml/L, 125.4ml/L, 125 and controls did not significantly ($p>0.05$) from each other. Relationship of seed mortality of red tilapia with concentrations of oil palm waste for 96 hours during the acute toxicity test was related of positively linearly (Figure 2).

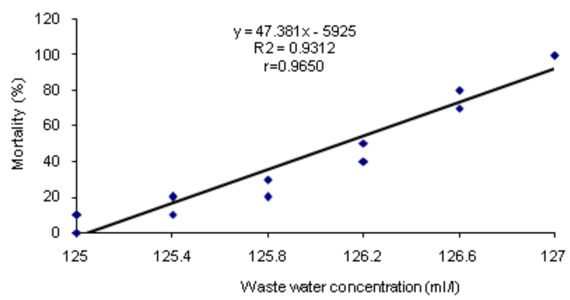

Figure 2: Tilapia seed mortality relationship with the concentration of industrial waste palm oil for acute toxicity tests

Figure 2 showed that the regression equation y = 47.38 x - 5925 with R^2 = 0.9312 and r = 0.97. Contribution of concentrations of oil palm industrial waste to changes in mortality of tilapia is 93.12%, and the others are influenced by other factors. The same that this research was found by Romi (2003) and Jumedi (2003), that the relationship of concentration of industrial waste palm oil on cork fish and grouper. Closeness of the relationship between the concentrations of palm oil effluent tilapia mortality may be caused by such waste toxic power at a certain level of concentration that can shut down the test organism. According to Said (1996), contain palm oil effluent has COD, BOD, suspended solids and emulsified high fat. Deposition and decomposition of organic matter that can gradually cause a decrease and an increase in pH resulting in deterioration of water quality causes the death of test organisms.

The values LC1-99 96 hours of liquid industrial waste of the oil palm seed tilapia in Table 2 and Figure 3.

Table 2: The values LC1-99 96 hours of liquid industrial waste of the oil palm seed tilapia (*Oreochromis* sp.)

LC (%)	Concentration (ml/L)
1	124, 97
5	125, 21
10	125, 36
15	125, 47
50	**126, 06**
85	126, 89
90	127, 13
95	127, 52
99	128, 39

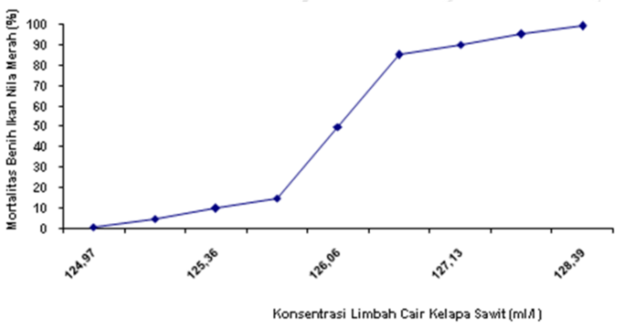

Figure 3: The values LC1-99 96 hours of liquid industrial waste of the oil palm seed tilapia (*Oreochromis* sp.)

Table 2 and Figure 3 shows the 96-hour LC50 value is 126.06 ml/L. This value indicates that if the palm oil industry wastewater into the waters with a concentration of 126.06 ml/L will cause the death of the seed of red tilapia (*Oreochromis* sp.) 50% for 96 hours.96-hour LC50 value of this study is ± 3 times greater than 96 hr LC50 values obtained by Romi (2003) (39.35 ml/L) with different organisms (fish larvae cork) of the same toxicant. This indicates that the liquid waste palm oil industry more toxic the cork fish larvae when compared with red tilapia seed. Koesumadinata and Sutrisno (1997) suggest that the sensitivity (susceptibility) organisms to toxicant are different according to the type and size of the organism.

Different waste concentrations provide a different form of response in each individual fish (Syafriadiman, 2000). Observations made during the acute toxicity test to seeds red tilapia (*Oreochromis* sp.) there are symptoms of behavior and morphology in normal conditions, sub lethal and lethal. Toxicant in this study may change the conditions of tilapia were initially normal to be lethal. Environmental disruption caused by industrial wastewater palm oil has caused the fish to become stressed, so it looks to be a different response depending on the sensitivity and resistance of fish. Said (1996) states that palm oil wastewater has potential as an environmental pollutant because of the smell, it contains the value of COD and BOD and suspended solids are high and oil in water emulsion. If sewage is discharged directly into the river portion will settle, decompose slowly, consume dissolved oxygen, causing turbidity, and issued a sharp odor. These factors are thought to cause stress tilapia seed, nearly dead (sub-lethal) and the occurrence of death, especially at high concentrations.

Under normal conditions of tilapia seed as test organisms in this study movement active, agile, balanced and Courant body morphology are not damaged. Under normal conditions of tilapia seed in this study with normal cork fish larvae in the study Romi (2003), i.e. no fins and scales are separated, fish eye clean and very responsive to stimuli. So also the fish's mouth opening and operculum move regularly and if the observed red gills.

Sub-lethal condition in this study that tilapia seed appears that movement is not balanced with direction uncertain, often spinning and blundering aquarium wall. Conditions begin to deteriorate fish body morphology characterized by the presence of scales apart. Opening the mouth and operculum fast moving fish and fish the less response to stimuli. While tilapia seed lethal condition during the study was not moving and paused at the base of the aquarium. Morphology characterized by the body breaks down the scales loose and easily removed from the body. Fish eyes and as if to stand out, while the mouth and operculum open. Fish have gills pale. Biological Safety Level of palm oil industrial wastewater for red tilapia seed was found during the study was 1.26 ml/L. Value of this study is Biological Safety Level of ± 3 times greater with the results of Romi (2003) for the same toxicant different test organisms, namely fish larvae cork (0.40 ml/L).

The results of the test sub-lethal

The growth of red tilapia during the study experienced an increase in P4 concentrations except 126.06 ml/L, where the seventh day all the fish died. Growth and survival of red tilapia seed during the study in Table 3.

Table 3: Growth and survival of red tilapia seed for 28 days of observation

P	U	Growth (g)					n_o	n_{28}
		0	7	14	21	28		
Po	1	3, 16	4, 36	4, 65	5, 42	6, 41	10	10
	2	3, 19	4, 37	4, 68	5, 53	6, 46	10	10
	3	3, 27	4, 44	4, 86	5, 51	6, 55	10	10
P1	1	2, 86	3, 35	3, 85	4, 37	5, 56	10	10
	2	3, 24	4, 34	4, 88	5, 62	6, 32	10	10
	3	3, 21	3, 29	4, 63	5, 45	6, 18	10	10
P2	1	2, 82	3, 57	3, 98	4, 42	5, 35	10	9
	2	2, 98	3, 60	4, 36	4, 98	5, 57	10	9
	3	2, 66	3, 05	3, 78	4, 62	5, 01	10	9
P3	1	2, 38	2, 68	3, 45	3, 91	4, 35	10	8
	2	2, 57	2, 96	3, 47	4, 06	4, 64	10	9
	3	2, 36	2, 67	3, 44	3, 90	4, 32	10	8
P4	1	3, 06	0	0	0	0	10	0
	2	2, 77	0	0	0	0	10	0
	3	2, 80	0	0	0	0	10	0

Description:

P: Treatment No: Total number of fish seed baseline
U: Replication N28: The number of fish seed after 28 days
Po: control P1: Concentration of oil palm waste 1.26ml/L
P2: Concentration of oil palm waste 31.52ml/L P3: Concentration of oil palm waste 63.03ml/L
P4: Concentration of oil palm waste 126.06ml/L

Table 3 showed that the seeds of red tilapia on the seventh day of the treatment concentration ofP4 (126.06 ml/L) died. It is likely the fish are notable to survive for seven days in a concentration of 126.06ml/L, due to the toxicity of waste palm oilcan be not tolerated by the red tilapia and eventually die. The study of the growth of the average weight of red tilapia seed for the growth of the test is the decline in the average growth of red tilapia seed weight with increasing concentration of palm oil effluent (Table 4).

Table 4: The growth of the absolute weight of seeds of red tilapia (*Oreochromis* sp.) during the growth test

Replication	Growth of the absolute weight (g)			
	Control	1, 26 ml/L	31, 52 ml/L	63, 03 ml/L
1	3, 25	2, 70	2, 53	1, 97
2	3, 27	3, 08	2, 59	2, 43
3	3, 28	2, 97	2, 35	2, 62
Total	9, 8	8, 75	7, 47	7, 02
Mean	3, 27	2, 92	2, 49	2, 34

Table 4 shows that the Mean growth in the absolute weight of red tilapia highest seeds contained in the control is equal to 3.27grams, and continued to decline until treatment with a concentration of 63.03ml/L of 2.34grams. The percentage decrease in the concentration of 1.26ml/L, 31.52ml/L, 63.03ml/L on consecutive controls were 10.70%, 23.85%, 28.44%. This decrease was probably caused by the poor condition of the water (Table 10) as well as the toxic material contained in the liquid waste palm oil. The relationship between the growths of the absolute weight of tilapia seed waste concentration is negative linear (Figure 4).

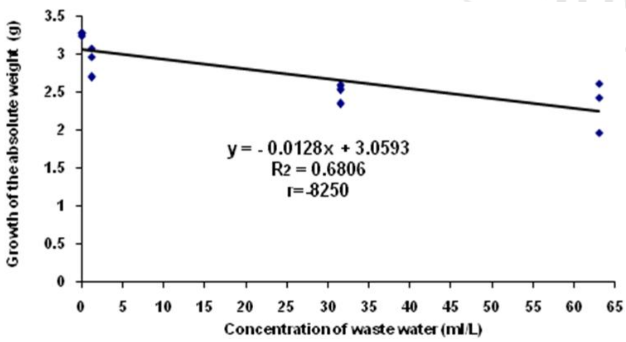

Figure 4: Relations absolute weight growth of red tilapia seed to the concentration of oil palm waste sub lethal test

Figure 4 shows the regression equation y=-0.0128x+3.0593 with a value of R2 =0.6806 and r=-0.8250. R2 values indicate the concentration of 68.06% contribution of oil palm waste to an absolute weight change in the growth of red tilapia seed during the study. Decrease in absolute weight growth was also found in the study Jumedi (2003). Absolute weight loss was caused by toxic materials that can affect the condition of the waters of the test fish. Based on the analysis of variance (Anova) that the concentration of waste oil not give a significant influence ($p<0.01$) on the growth of red tilapia seed absolute weight (*Oreochromis* sp.). As per the provision of test results anava concentration of palm oil waste at different exposure time of 96 hours gives highly significant effect ($p<0.01$) on mortality of red tilapia seed. Newman-Keuls test result that absolute control treatment had different weights highly significant ($p <0.01$) of the treatment concentration 31.52ml/L and treated with a concentration of 63.03ml/L, did not differ by treatment with 1.26ml/L. Treatment concentration of 1.26ml/L had different ($p<0.05$) between treatment concentration of 31.52ml/L to 63.03ml/L. While the control treatment did not differ ($p>0.05$) by treatment with concentration 1.26ml/L.

Value of daily growth rate of red tilapia seed was found to decrease with increasing concentration of palm oil effluent. Where the higher concentrations of the lower palm oil waste daily growth rate of red tilapia seed (Table 5).

Table 5: Daily Growth Rate of Seeds Red Tilapia (*Oreochromis* sp.)

Replication	Daily Growth Rate (%)			
	Control	1, 26ml/L	31, 52ml/L	63, 03ml/L
1	2, 56	2.40	2, 31	2.18
2	2.55	2.42	2, 26	2, 13
3	2, 51	2.37	2, 29	2.18
Total	7, 62	7, 19	6, 86	6, 49
Mean	2, 54	2, 40	2, 29	2, 16

Table 5 shows that the average daily growth rate of red tilapia seed in controls was 2.54%, on treatment with a concentration of 1.26 ml/L at 2.40%, on treatment with a concentration of 31.6 ml/L of 2, 29%, and the treatment with a concentration of 63.1 ml/L of 2.16%. The presence of this daily growth rate decreased; probably due to the water conditions are not conducive to the growth of red tilapia seed (Table 5). The relationship of the daily growth rate of red tilapia seed against palm oil effluent concentration is negative linear (Fig.5)

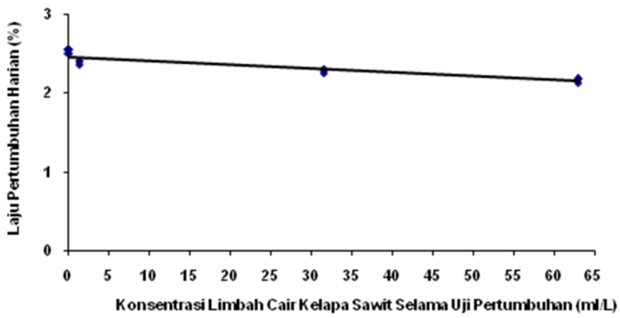

Figure 5: Daily growth rate relationship to the concentration of oil palm waste during the study

Figure 5 shows the regression equation y = - 0.005 x + 2.4666 with a value of R2 = 0.8487 and r = -0.9213. Based on the regression equation is then an increased concentration of one unit, it will reduce red tilapia daily growth rate of 0.005 times. Of the value of R2 is known that the concentration of oil palm waste contribution to the change rate of daily growth of red tilapia seed at 84.87%, while the rest is influenced by other factors with a strong negative relationship closeness (r = - 0.9213). Decrease in daily growth rate was also found in the study Jumedi (2003). Absolute weight loss was caused by toxic materials that can affect the condition of the waters of the test fish.

Based on the analysis of variance (Anova) it was found that there was a significant influence ($p < 0.01$) between the concentration of palm oil effluent to the daily growth rate of red tilapia seed (*Orechromis* sp). Then further test Newman - Keuls explained that the control treatment; 1.26 ml/L, 31.52 ml/L and 63.03 ml/L daily growth rate differences are very real to each other.

Water quality parameters

Values of water quality parameters during the study was almost the same in each treatment, as temperatures range between 26-29, pH 5-8, 1.2 to 6.4 ppm DO, turbidity 0-65 NTU and Ammonia from 0.04 to 0.840 ppm. During the study, the temperature ranges between 26-29 0C. Temperature range is still quite good for the growth of fish. In accordance explained Arie (2000), the optimal temperature for growth of fish in the range of 25-30 0C. Then Boyd (1979) also suggests that tropical areas the temperature range 25-320C is still viable for the growth of aquatic organisms.

pH during the study ranged between 5-8. PH values found 5 result of water mixing with sewage pH 6 pH 4, pH 5 due registered only at the beginning of the study. Fluctuations in pH in this study also occurred in the study Romi (2003) with the same toxicant, the waste oil with different types of fish, i.e. fish larvae cork. PH range is still good for fish life. According to Brown (1980), water pH less than 4 or greater than 11 can cause death in fish. While the ideal pH for the life of aquatic organisms ranged from 6.5 to 8.5 (Pescod, 1973).

DO recorded in the control treatment from 6 to 6.6 ppm and the concentration of 1.26 ml/L range between 5-6 ppm and the DO concentration decreases with increasing palm oil waste so that the concentration of 126.06 ml/L. Apparent decline in oxygen concentration associated with the concentration of a given oil palm waste, because waste palm oil has a high organic matter. The low value of dissolved oxygen in this study led to high mortality of seeds of red tilapia (*Oreochromis* sp) up to 100%.

High values of ammonia also cause high mortality. Ammonia range recorded during the study was 0.04 to 0.840 ppm. Boyd (1979) states sandstone toxic than NH_3 for short-term exposure period is 0.6 to 2 ppm. Afrianto and Liviawaty (1992) states that low concentrations of ammonia can cause tissue damage gills, making fish hard to take oxygen from the environment.

4. Conclusion

The results showed that palm oil waste was highly significant ($p < 0.01$) on seed mortality tilapia (*Oreochromis* sp.) 96-hour LC50 values for toxicity studies was 126.06 ml/L, with the safe biological limit values (Biological Safety Level) is equal to 1.26 ml/L. The test results and the growth in the absolute weight daily growth rate for 28 days showed a highly significant difference in effect ($p < 0.01$) against concentration of palm oil effluent. The higher concentration of palm oil effluent given the smaller growth of absolute weight and daily growth rate of seeds of red tilapia (*Oreochromis* sp.). Water quality parameter values for toxicity testing and test growth in the bad with the increasing concentration of palm oil effluent.

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