

## Assessing The Effectiveness of Biological, Chemical and Physics Treatment for Reducing Formalin in Sea Water

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### Abstract

Formalin is frequently found in water that has been used for the cultivation of grouper fish. If it enters public waters, the leftover water is feared to disturb organisms living in it. The objective of this research is to reduce formalin concentration in sea water through five treatment and control (no treatment), which is aeration, UV light from 10-watt light bulb, 3 g l<sup>-1</sup> addition of active charcoal from corncob, 3 g l<sup>-1</sup> addition of yeast, 10 ml l<sup>-1</sup> addition of bacterial community. The formalin concentration was observed every 24 hours using formalin test kit for four days (4x24 hours). The results are that bacterial community treatment reduces formalin concentration to 0 mg l<sup>-1</sup> after 48 hours, yeast treatment reduces the concentration to 0 mg l<sup>-1</sup> after 96 hours, aeration reduces the concentration to 5 mg l<sup>-1</sup> after 72 hours yet bouncing back up after 96 hours. Control, UV treatment, and active charcoal treatment are similar in that those three treatments do not reduce the formalin concentration, remaining at 10 mg l<sup>-1</sup>. Therefore, addition of bacterial community can reduce formalin until 0 mg l<sup>-1</sup> concentration in two days, and yeast addition can reduce it until 0 mg l<sup>-1</sup> in four days. Hence, further researches about dissipating formalin in sea water using bacterial community with different dosage and time length are required.

Keywords: formalin, treatment, sea water

### INTRODUCTION

One of the obstacles in cultivating groupers is disease, particularly parasite. There are 35 different parasites that can attack groupers [1]. Most parasites that attack groupers are worms and leeches from the class of Monogenean, Trematode, and Hirudinea. Those parasites have suckers enabling them to stick on the fish's body, and they are difficult to remove [1]. Several types of worm that attack the outer body of groupers are Trematodes and Monogeneans such as *Benedenia*, *Neobenedenia*, *Diplectanum*, *Haliotrema*, *Calligus* sp, *Rhexanella* sp of Crustacean, Microcotylid of Monogenean [2], and leeches (Hirudinea) of Annelida [2] [3].

The inner body of groupers is also vulnerable. *Prosorhynchus luzonicus* from Trematoda Digenea class is found in the fish's stomach, *Gorgorhynchoides golvani* from Acanthocephala class is found in the fish's gut and body cavity, and *Allonematobothrium epinepheli* of Digenea class is found under the gill cover [3].

To overcome the parasite problems, grouper cultivators use formalin (the structural formula is depicted in Figure 1, which is frequently used to soak fish infected by parasites and fungi [4] [5] [6] [7]. A soaking process in 100 mg l<sup>-1</sup> solution for one hour is effective for removing parasites in fish [8].

Figure 1. Structural Formula of Formalin

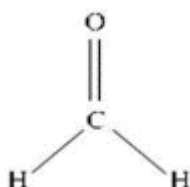
According to the decree of Ministry of Marine and Fisheries number 52 of 2014 regarding drug classification, formalin is a limited non-prescription drug, which means that it is a prescription drug, and, for fish, its usage should be accompanied with a specific warning sign, and its amount, dosage, form, and special usage are regulated. Formalin should not be disposed directly to the environment because it is actually a prescription drug. Fish cultivators usually dispose water containing formalin that was used to soak fish straight to public waters due to the absence of either waste processing installation or simple and proper method to treat the formalin-

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polluted water. Its because that formalin is a sensitizing agent that can cause an immune system response upon initial exposure for human. Formalin is also known as human carcinogen. Prolonged exposure to formaldehyde has been associated with cancers of the lung, nasopharynx, oropharynx and nasal passages (nose and throat) and some studies suggest formaldehyde may cause leukemia, particularly myeloid leukemia, in humans [9].

Waste disposal of formalin in the public waters can kill phytoplankton, benthic and other organism with high sensitivity of formalin [10]. Its can be affect of food chain in the ecosystem. Formalin did not accumulate in the seawater, but it take little time for disappear [10].

The waste management especially formalin can be use physically, chemically and biologically. In chemical processing it will occur from a substance to another, as well as formalin. This research on Chemically process is by adding air (oxygen) and activated charcoal, so that it can change the chemical form of formalin into a harmless material. Whereas for the physical processing, UV irradiation is carried out. Irradiation technology can produce ionization in a material and then initiate a chemical reaction so that it can break its bonds [11]. In this study UV light was chosen because UV lamps were easy to obtain. The biological waste management process is carried out by utilizing the activity of organisms to remove pollutants [12]. In this study yeast and bacterial community were used. All treatments use materials and tools that are easily available and affordable.

The aim of this research are to determine simple treatments to reduce or even dissipate formalin content in seawater and to identify the shortest time of each treatment in reducing formalin concentration. Thus, researches on the treatment of the polluted water are required, so the disposed water is safe for public water and microorganism.

#### **MATERIAL AND METHOD**

This research uses completely randomized factorial design, where two variables to be measures are type of treatment and spent to decompose formalin waste in grouper soaking. Cultivated wastes disposed of in public waters should be safe from formalin. Therefore is need for treatment to reduce it. Treatments are used from materials or tools that are simple and easy to obtain. The treatments are aeration, UV, active charcoal, yeast, bacterial community, and

one control (non-treated sea water). The dosage of the formalin is  $100 \text{ mg l}^{-1}$  with the treatment length of 24, 28, 72, and 96 hours .

Materials used in this research are formaldehyde 37%, sterilized sea water with the salinity of  $25 \text{ g l}^{-1}$ , corncob active charcoal, yeast, bacterial community, and formalin detection kit from Merck with series number HC731983. The bacterial community used is EM4 (Effective Microorganism 4) which contains fermented bacteria, fermented fungi, actinomycetes, photosynthetic bacteria, phosphate solvent bacteria and yeast.



Figure 2. Formalin Detection Kit



Figure 3. Bacterial Community

The tools used in this research are jars, aeration kit, UV lamp, measuring cylinders, sample bottles, dropping pipettes, gloves, and medical face masks. After the formalin concentration measurement results are obtained, completely randomized factorial design is used to identify the relationship between two factors, i.e. type of treatment and length of time.

#### **RESULT AND DISCUSSION**

Formalin in its purest form is colorless, highly toxic, highly flammable, and pungent gas, but it contains several methanol as stabilizers. Paraformaldehyde is a white crystal powder from polymerized formaldehyde, a flammable solid that can produce formaldehyde gas when heated or mixed with water. Many laboratories at University of Washington use formalin solution and paraformaldehyde solution and solid as parts of their research and clinical activities. It is commonly used in tissue repair and conservation and as organic chemical reagent. The most frequently used chemical material at University Of Washington is 10% neutral buffered formalin, which contains 4% formaldehyde [9].

Formalin is usually used as disinfectant and fixative (BG/L, 2014). According to the Agency of Drug and Food Control of Republic of Indonesia (BPOM), the use of formalin in food is very dangerous. It has both short and long-term effect, depending on its exposure to the body. The effects are respiratory irritation, regurgitation, dizziness, burning throat, body temperature drop, and itchy chest. In addition, it can damage liver, heart, brain, spleen, pancreas, central nervous system, and kidney.

Formalin soaking is used to treat parasitic disease [4] [5] [6] [7]. The use of 100 mg l<sup>-1</sup> of formalin for one hour soaking of grouper is quite effective to remove monogenean and digenean parasites (Slamet *et al*, 2008). The waste of the soaking process is usually disposed directly to the waters, which is dangerous for the land and water environment around the cultivation area. Although ambient seawater area nearby discharged effluents from land-based tank or net cage of marine finfish is not stagnant seawater body, partial inhibition or kill of marine organisms showing sensitivity to formalin may occur. Although formaldehyde did not accumulate in seawater, it took a little time to disappear. Discharged formalin from marine finfish farm in land-based or net cage may affect acute toxicity to phytoplanktons used for feed of fish and shellfish [10].

Significant result variations were found from the treatments given during this research; some reduce the concentration, and some others completely dissipate it. The result of DMRT shows that control, UV, and charcoal treatments do not give any significant difference, while aeration, yeast, and EM4 treatments do. In terms of treatment's length of time, the best result is after 96 hours, while the calculation results from 48 and 72 hours of treatment are not significantly different.

The measurement results show decreases of formalin concentration in the sea water with the salinity of 25 g l<sup>-1</sup> at the temperature between 25 and 26 degree Celsius. The concentration decreases are different for each treatment in each time length. The detail of the mean decrease of formalin concentration is depicted in Figure 4.

The first treatment is aeration. The measurement result shows that the formalin concentration decreases to 20 mg l<sup>-1</sup> after 24 hours of treatment and to 10 mg l<sup>-1</sup> after 48 hours of treatment for every repetition. However, different measurement results occur

on the third day, in which a decrease to 5 mg l<sup>-1</sup> took place in the third repetition, but there was no concentration decrease in the first and second repetition.

Different measurement result between repetition also took place after 96 hours, in which there was no difference between the first repetition the day with the first repetition on the previous day, yet the concentration increased to 20 mg l<sup>-1</sup>. This happened due to condensation of water vapor in the jar. The formalin concentration measurement result after aeration is available in Figure 5.

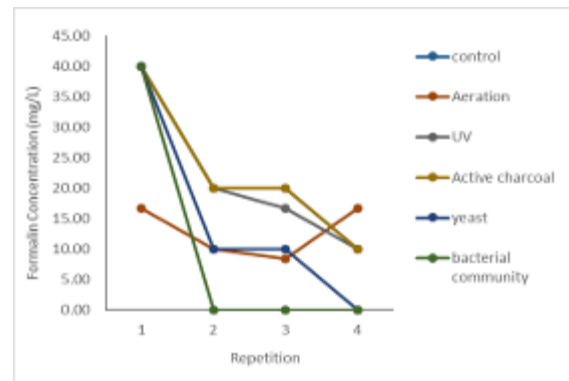


Figure 4. The Chart of the Mean Results of Formalin Measurement

Chemical oxidation is by adding oxygen (aeration) to sea water. At this time the oxidation-reduction reaction is expected, where formalin will undergo a reduction reaction and the oxidizing agent will be reduced. This is expected to be able to decipher the chemical bonds of formalin into materials that are harmless and safe to dispose of into public waters. Metcalf, et al (20003) in Saptaati, *et al* (2018) said that the addition of oxygen to liquid waste can decompose chemicals by reaction:  $O_2 + 4H^+ + 4e^- \leftrightarrow 2H_2O$  with potential oxidation values +1,23.

With positive oxidation values, formalin should be able to decompose with the addition of oxygen (aeration). However, the measurement of formalin levels showed that there was no significant difference between the treatment and controls, so it can be seen that the addition of oxygen cannot reduce the level of formalin in seawater. Saptaati, *et al* (2018) [12] explained that the use of aeration in pollutant water is often limited by the slow reaction rate, so it is need for additional catalysts. Therefore the addition of aeration to the decrease in formalin levels still cannot work optimally.

Besides that aeration does not reduce formalin concentration in sea water. This finding supports the research of Hua, Z., et al (2018) [13]. The research mentioned that the absence of DO at pH 7 inhibits the degradation of PPCPs (Pharmaceuticals and Personal Care Products). It also mentioned that the absence of DO prevents ozone generation that produces degradation pressures form sever PPCPs with high ozone reactivity, but it did not mention the presence of formalin, so further researches to confirm it are necessary.

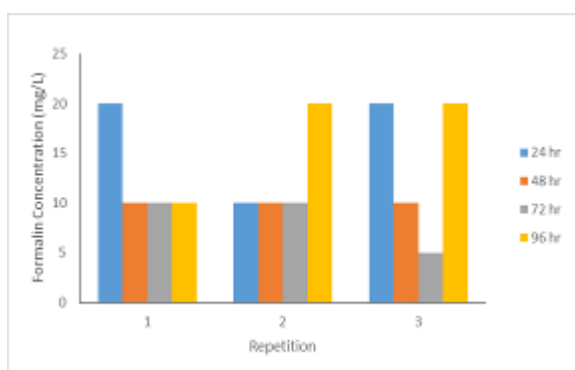


Figure 5. The chart of Formalin Concentration Measurement for Aeration Treatment

The next treatments UV, and active charcoal. The measurement results show that there is no significant difference between the treatments in decomposing formalin in sea water. However, the treatment's length of time significantly influences the decrease of formalin concentration in sea water. The measurement results of those treatments with different length of time is presented in Figure 4.

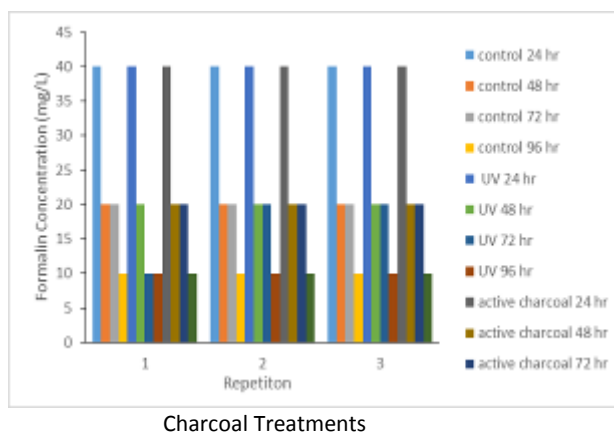
UV irradiation is carried out with a closed container without any incoming light other than itself. This is use to make UV irradiation effective without other influences. UV light is used as a water filter for drinking and cultivation. Because it can damage the cell, it is expected that UV irradiation can decipher formalin chemical bonds. However, the results of the study show different things. With the presence of UV irradiation the levels of formalin in sea water are not significantly different from the control.

UV irradiation treatment apparently is not significantly different from control; it did not reduce formalin concentration in sea water, because UV irradiation cannot be used to decompose chemical compound. This is similar to the research of Mansouri et al. (2018) [14] that UV lighting is not effective in the photolysis of DEP (diethyl phthalate). The research explained

that the maximum wave length during the UV lighting is higher than the wavelength of absorbs DEP. It also applies for formalin, that UV lighting does not reduce the formalin concentration in sea water.

Active charcoal was used with the expectation that it will absorb formalin. Charcoal is any kind of carbon, which has high absorption for gas and water vapor as well as solid colloid [15]. This research used activated carbon made from corncob. Activated carbon is a material that is often used in water filtration. Its ability to absorb oganic and inorganic compounds which are toxic causes active carbon to be suitable as a treatment in reducing formalin levels. In this study, activated carbon from corn cobs in the form of powder was used.

Figure 6. The chart of Formalin Concentration Measurement for Control, UV, and Active



The characteristics of activated carbon as adsorbent are very influential on the adsorbtion process. The addition of activated carbon is expected to be able to absorb the formalin contained in seawater, so that it can reduce formalin levels. The result is that there is no significant different between control and activated carbon treatment (Figure 4), which means that activated charcoal does not reduce formalin concentration in sea water. According to Hutami (2006) [15], activated charcoal help absorb poisonous compound in sterilized media. Nevertheless, the result of this study shows that activated charcoal cannot absorb formalin in sea water.

Active carbon should be activated with other compounds such as  $\text{NH}_4\text{HCO}_3$  so the absorbtion

Figure 7.  $\text{NH}_4\text{CO}_3$  Reaction plus Carbon of formalin can be optimized, as Fig 7. Activation will cause carbon to be polar so that it can adsorb polar formaldehyde [16].

Figure 4 shows that yeast can reduce formalin concentration in sea water after the fourth day or 96 hours. Yeast changes formalin into formic acid. Therefore, the addition of yeast is followed by addition of molasse or sugar as the media for fungi. According to Nur Hidayat (2018) [17], yeast and fungi grow well at the temperature of 24 to 46 degree Celsius, similar to the temperature during this research, i.e. 25 to 26 degree Celsius. Therefore, yeast can work optimally during the fermentation, so it can reduce the formalin concentration in sea water.

To survive, yeast needs water, food and an appropriate environment. These single-celled bacteria will work easily when added to sugar and warm temperature conditions. During this study it was found that the addition of yeast can transform formalin into formic acid. It can be seen from the odor after 4 days. However, to do so, it takes time (4 days) because of the adaptation to salinity 25 ppt. So after 4 days, the yeast can be removed and turned into formic acid.

Bacterial community dissipate formalin in sea water in two days due to the metabolic process of the bacteria in the community. The activities of microorganisms change environmental condition, so population components and their habitat also changed by taking components available in their environment, microorganisms will proceed their metabolism [17]. This caused the decrease in formalin concentration in waters because it is used in the metabolism of microorganism in the bacterial community [18]. One of the bacteria that is included in the bacterial community is lactobacillus, which is a bacterium frequently used as probiotics. It produces lactic acid during its metabolism. The enzyme produced by this bacterium is anaerobic, so it does not tolerate oxygen [17]. Hence, this bacterial community addition must be done in limited media with or without oxygen. Therefore, the increase of the bacterial community should be done in closed media with little or no oxygen. This is relevant with the activities during the research.

## CONCLUSION

The result of this research it was concluded that the use of UV, activated charcoal and aeration was not significantly different from the control in the sense that the use of these three treatment was not effective in reducing formalin. Biological treatment with the use of yeast can reduce formalin levels from 100 mg l<sup>-1</sup> to 0 mg l<sup>-1</sup>

within 4 days, while EM4 can reduce formalin levels from 100 mg l<sup>-1</sup> to 0 mg l<sup>-1</sup> within 2 days.

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