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Correlation between BOD/COD Ratio and Octanol/Water Partition Coefficient for Mixture Organic Compounds

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Abstract

Correlation between the BOD/COD ratio and Partition coefficient of octanol/water (Pow) on a single organic substance shows that the Pow value is directly proportional to the toxicity level and inversely proportional to BOD/COD ratio. This research examined the correlation to a mixture of organic substances. The objective is to obtain a varied range of substances, as well as determining the quality of wastewater discharging to fresh waters. Need for analysis of organic substances used as antiseptics during the Covid-19 pandemic. In addition, organic substances from the organophosphate pesticide class, diazinon, were used. BOD5, COD, Pow, and LC50-96h toxicity tests using Daphnia magna were used. Six types of the mixture of organic substances included diazinon-formaldehyde-isopropyl alcohol, ethanol-oxalic acid-formaldehyde, isopropyl alcohol-glycerol-lactose, acetic acid-isopropyl alcohol-formaldehyde, sucrose-glycerol-acetic acid, and oxalic acid-formaldehyde-diazinon, with 3 different concentrations of 10, 100, and 1000 mg/L, three repetitions. The lowest BOD/COD ratio (<0.2) and the highest Pow value (>4) are found in diazinon-formaldehyde-IPA. Its toxicity in D. magna also showed the lowest LC-50 (11.82 mg/L). Whereas, sucrose-glycerol-acetic acid had the highest BOD/COD ratio (>0.7) and lowest Pow (<0.7) with the highest LC-50 (567.88 mg/L). Other organic substances mixtures have characteristics in the range of these mixtures. Pow variability and the BOD/COD ratio have a negative correlation. A mixture of organic matter is more biodegradable making it has a higher tendency to dissolve in water.

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1. Introduction

In accordance with the quantity complexity, organic substances have the complexity of pollutant quality[1,2]. During the Covid-19 pandemic, there are several organic substances used as Covid-19 antiseptic substances, namely glycerol, ethanol, and isopropyl alcohol[3]. In addition, organic substances from the organophosphate pesticide class, diazinon, are also used. Diazinon is favored by farmers because of its faster and more significant way to eradicate pests than other pesticides[4-6].

The organic substances used in this study were a mixture of glycerol, ethanol, isopropyl alcohol, diazinon, and five other substances: sucrose, oxalic acid, acetic acid, formal dehyde, and lactose. These organic substances are often used so that a lot of them are wasted in the environment. Sucrose in the manufacture of food products functions as a sweetener and preservative[7]. Lactose is found in milk (35-45%)[8]. Formaldehyde is found in household cleaners, paper, shampoos, deodorants, toothpaste, lipsticks, nail dyes, and pesticides[9,10] Oxalic acid is used in textile manufacturing and metal surface treatment and leather tanning[11,12]. Acetic acid is most widely consumed and traded in industry and laboratories, known for providing sour taste and aroma in food[13,14].

To determine the long-range ratio of 6 types of mixtures organic substances used BOD_5, COD and Pow analysis. The BOD/COD ratio represents the biodegradability of wastewater, where the ratio is in line with its bio degradability[15,16]. A substance with a tendency to obtain bio concentration related to lipophilic properties and their persistence were analyzed by the Pow coefficient[17,18]. Pow analysis can determine the tendency of a substance, whether to biota or water, which describes the hydrophobic and hydrophilic properties of aquatic organic[19,20].

Waste water with a high content of pollutants can be harmful to the biota both in salt water and fresh water. It is necessary to verify laboratory results with a toxicity test to determine the quality of the disposal. In this research, the LC50-96 h toxicity test was carried out using *D. magna* which is the representation of freshwater organisms due to its distinctive characteristics among others, such as has an even distribution throughout the year, has economic and ecological values, is easy to culture in the laboratory, is free from parasites or diseases and is in accordance with the interests of the test[21,22]. *D. magna* is very sensitive to environmental changes so that it can determine the level of waste that can cause toxic effects as an aspect of monitoring water quality and water pollution (early warning system)[23-25].

The novelty of this study is that the 6 types of organic substances involved have a very varied range of materiality, including the quality status of the mixture. This makes the mathematical model of the BOD/COD and Pow correlation better. In addition, the need for analysis of organic substances used as antiseptics during the Covid-19 pandemic was also fulfilled. The values of BOD_5, COD, Pow and the LC50-96h toxicity test using *D. magna* were also examined.

2. Materials and Methods

2.1 Study Area

This research was conducted on a laboratory scale in the Environmental Quality Management Laboratory and Water Treatment Technology Laboratory, Environmental Engineering Department, Institut Teknologi Sepuluh Nopember, Surabaya.

2.2 Main Research

The main research stage was carried out using BOD analysis with the Winkler method[26,27]. The principle was using iodometric titration. The wastewater sample was first added with a solution of $MnCl_2$ and NaOH-KI (Merck, Germany) so that MnO_2 deposits occurred. By adding H_2SO_4 (Merck, Germany), the precipitate that occurred would dissolve again and also liberate iodine (I_2) molecules which were equivalent to dissolved oxygen. The liberated iodine was then titrated with a standard solution of sodium thiosulfate ($Na_2S_2O_3$) (Merck, Germany), using the starch solution indicator (starch). The next research stage was COD analysis with closed reflux spectrophotometric[28,29]. Organic compound was oxidized by $Cr_2O_7^{2-}$in closed reflux to give Cr^{3+}. The amount of oxidant required was expressed in terms of oxygen equivalent (O_2 mg/L) measured by visible light spectrophotometry. $Cr_2O_7^{2-}$ strongly absorbs at a wavelength of 420 nm and Cr^{3+} has a strong absorption at a wavelength of 600 nm. In the test sample with a higher COD value, dilution was carried out first before

testing. For COD values less than or equal to 90 mg/L, the decrease in $Cr_2O_7^{2-}$ determined concentration was at a wavelength of 420 nm.

The next research stage was Pow analysis using gas chromatography[30,31]. A number of mixed substances (for example formaldehyde-ethanol-oxalic acid) are dissolved into a mixed solution of octanol (5 mL) and water (5 mL) in test tubes. The test tubes were shaken using a shaker for 24 hours for homogeneous mixing. Furthermore, the solution was separated using a centrifuge. Water samples could be taken for the content analysis of substance (for example obtained c) and the Pow results were obtained from gas chromatography (GC7900, Hongkong).

2.3 Toxicity Test LC_{50}-96 h on *Daphnia magna*

The method of toxicity testing for *D. magna* was described by the USEPA in 2002 for Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms[32]. The age of *D. magna* used was less than 24 hours. Room temperature was maintained at 22-31°C. The pH, dissolved oxygen (DO), and temperature were being kept under observation every day. A total of 10 *D. magna* were put into various concentrations (10, 100, 1000 mg/L). Furthermore, observations were made on the number of immobile *D. magna* at 24, 48, 72, and 96 hours. The LC50 value was then calculated using probit repeated three times analysis[33-35].

3. Results and Discussion

3.1 Correlation between BOD/COD Ratio and Pow

The BOD and COD analysis showed that the characteristic of the substances for acetic acid-IPA-formaldehyde, sucrose-glycerol-acetic, and IPA-glycerol-lactose acid is biodegradable (Table 1). The order of the largest biodegradability level is sucrose-glycerol-acetic acid, organic IPA-glycerol-lactose, and acetic acid-IPA-formaldehyde. The characteristic of the organic mixture of diazinon-formaldehyde-IPA, formaldehyde-ethanol-oxalic acid, and oxalic acid-formaldehyde-diazinon is non-biodegradable. Organic pollutants that are difficult to decompose /non-biodegradable are indicated by very low BOD/COD ratio values (<0.02).

The results showed a mixture of organic substances with the lowest BOD/COD ratio, namely diazinon-formaldehyde-IPA, which was 0.144 at a concentration of 1000 mg/L. The entry of diazinon into the waters can decrease water quality in terms of physics (temperature and brightness), chemistry (pH, carbon dioxide, and dissolved oxygen), and biology (fish, plankton, macrophages, and benthos) which will result in no balanced aquatic environment[36,37]. Formaldehyde can reduce the activity of microorganisms because it is an accumulative and strong reducing agent[38,39]. The mixture of organic substances with the largest BOD/COD ratio is

Table 1. LC_{50}-96h value of mixed organic compounds against *Daphnia magna*

Mixed Organic Compounds	*LC_{50} value* (mg/l)	*Estimated Probit Line*
Diazinone-formaldehyde-IPA	11,82	m=1,363x+3,538
Formaldehyde-ethanol-oxalic acid	46,69	m=2,452x+0,907
IPA-glycerol-lactose	278,39	m=1,527x+1,267
Acetic acid-IPA-formaldehyde	205,09	m=1,715x+1,035
Oxalic acid-formaldehyde-diazinone	43,67	m=2,479x+0,934
Sucrose-glycerol-acetic acid	567,88	m=1,469x+0,954

m = mortality of the test organism and x = concentration of mixed organic compounds

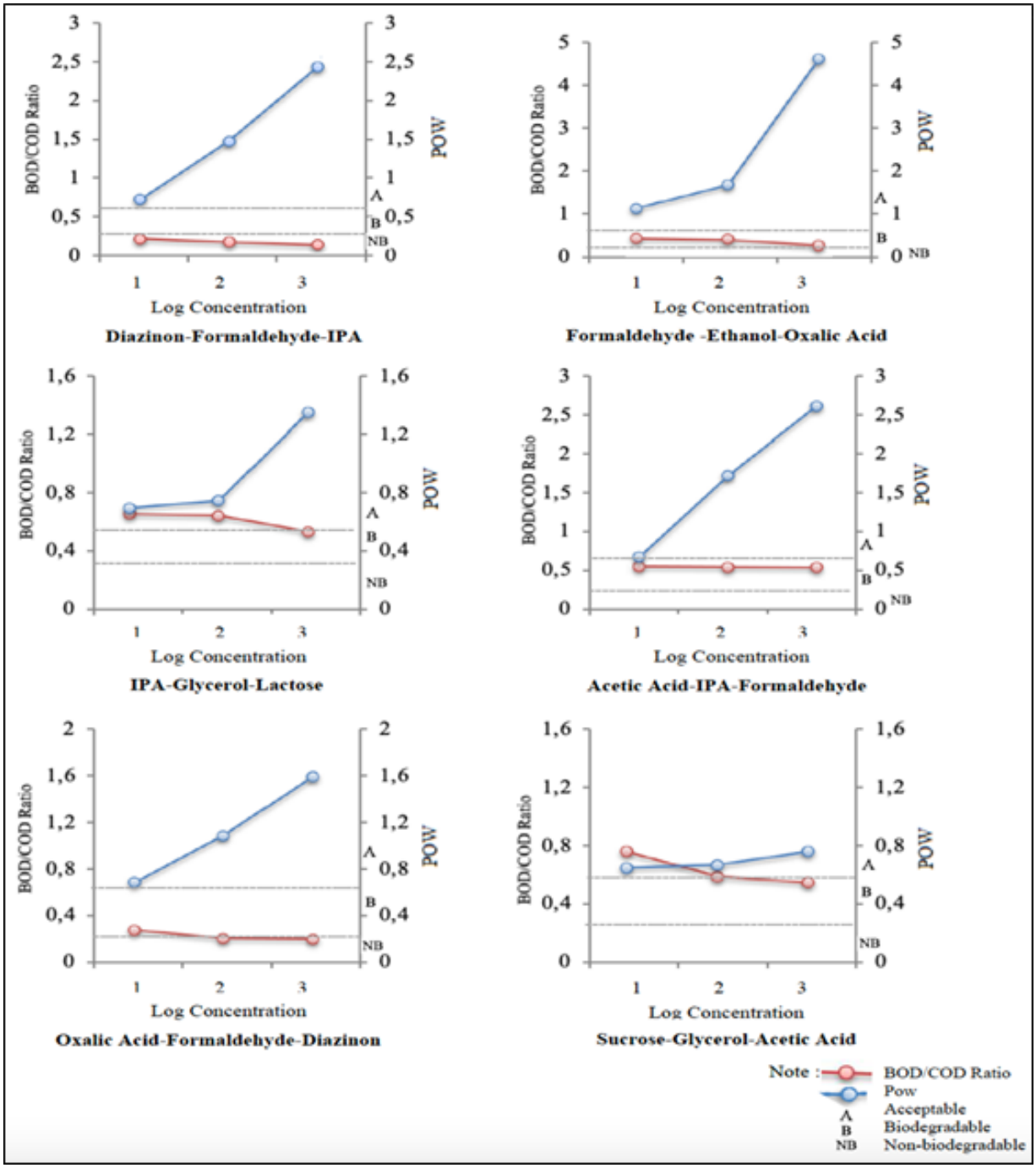


Figure 1. Biodegradability of mixed organic compounds.

sucrose-glycerol-acetic acid of 0.769 at a concentration of 10 mg/L (biodegradable). The nature of glycerol which is easy to absorb water has an energy content and a high water solubility index, which is a source of carbon for microorganisms[40,41] (Figure 1).

From the results, the Pow value shows a tendency towards biomass. A mixture of non-biodegradable organic substances, for instance, diazinon-formaldehyde-IPA, formaldehyde-ethanol-oxalic acid, and oxalic-formaldehyde-diazinon are more focused on biomass.

Due to the average POW value is >1, which is higher than to a mixture of biodegradable organic substances, namely IPA-glycerol-lactose, acetic acid-IPA-formaldehyde, and sucrose-glycerol-acetic acid. The more non-biodegradable a substance, the greater it is tendency to produce biomass. This is because only a small part of the substance can be dissolved in water, urine, or other waste systems. A person consumes a lot of formal dehyde, it will accumulate in the body, reducing excretion compared to sucrose. The Pow value of a mixture will be higher along with the level of toxicity. Mean while, the smaller the BOD/COD ratio, the more toxic the concentration. The correlation between Pow and BOD/COD ratio was found to be negatively correlated in the range of 0.1 - 0.9 for 6 types of mixtures of organic substances. A mixture of organic substances that are more biodegradable will tend to dissolve in water.

3.2 Toxicity Test LC_{50}-96 h on *Daphnia magna*

The LC50-96 value of a mixture of biodegradable organic matter is greater than that of a mixture of non-biodegradable organic matter (Table 1). This means that a mixture of non-biodegradable is more toxic than a mixture of biodegradable organic substances and has a more tendency towards biomass. Based on observations, the LC_{50}-96 hour value of diazinon-formaldehyde-IPA against *D. magna* was obtained at 11.82 mg/L (Table 1). The response of *D. magna* to the concentration of the diazinon-formaldehyde-IPA showed the highest mortality.

The probit line equation obtained from the LC50 toxicity test is m=1.363x + 3.538, where *m* is the dependent variable showing the mortality of the tested animals for 96 hours. The value of a (3.538) is the y-intercept of the value and b (1.363) is the slope of the regression line, x is the concentration of a mixture of organic substances. The higher the concentration of the organic matter mixtures the higher the mortality rate of the test animals. This is indicated by a positive regression coefficient value. The greater the slope value, the higher the level of toxicity (small LC50 value) (Figure 2).

Based on the observations, after 24 hours of exposure clinical symptoms due to diazinon-formaldehyde-IPA appear in *D. magna*, especially at concentrations of 100 and 1000 mg/L. *D. magna* swims irregularly and subsequently dies. This symptom according to[42-44] is a response that occurs when the cell or sub cellular processes in living things are disturbed by a substance to the extent that it causes immediate death. According to[45,46], toxic exposure of *D. magna* can be identified from its behavior, namely by hyperactive movements, floundering, and paralysis. This is a way to reduce toxic biochemical processes in the body so that it can slow down the death.

The pH of the test solution in this research ranged from 7.1 – 8.3. It is in the recommended pH range for the

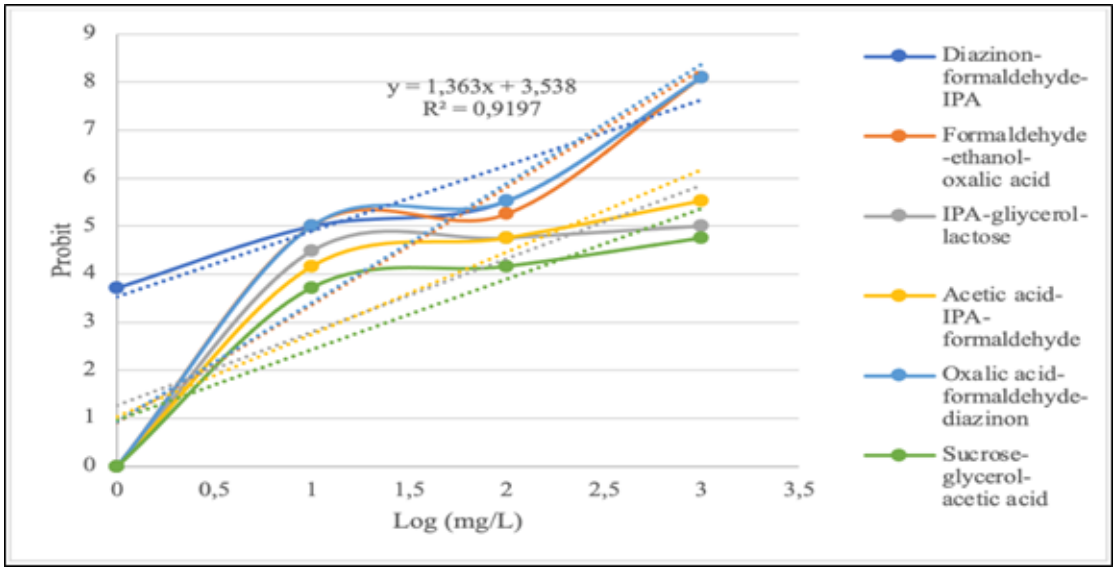


Figure 2. *Daphnia magna* probit line equation.

optimal life cycle of *D. magna*, which is 7.2 – 8.5[47,48]. DO of the test solution ranged from 5.0 – 6.6 mg/L. This DO range is above the recommended DO in the optimal life cycle of *D. magna*, which is > 3 mg/L[49,50]. This condition is very good for *D. magna* because DO or dissolved oxygen is needed by *D. magna* in metabolic processes in its body. If DO levels in the water are not available in sufficient quantities, it will cause the water quality to decline and can lead to the death of microorganisms. The temperature of the test solution ranges from 27.6°C – 28.8°C. It is in the optimal temperature range on the life cycle of *D. magna*, which is 22°C – 31°C[51,52].

These observational data indicate that the pH, DO, and water temperature during the test were still within the recommended ranges for *D. magna* life. The condition of those three aspects is maintained so that *D. magna* can live optimally. It can be concluded that the death of *D. magna* was caused by contaminants in the test solution. In the control solution, there were no physical symptoms due to poisoning and no dead *D. magna* found during the 96-hour observation time. This indicates that the quality of the maintenance media during the test is in good condition. Meanwhile, the other samples showed physical symptoms at the time of initial exposure to the test solution. Test animals move hyperactively until after a while there is no movement anymore. *D. magna* that has died turns white and sinks to the bottom of the container.

Test animals exposed to toxic contaminants show signs of moving hyperactively, floundering, paralyzing, and then going dead[53,54]. Clinically, test animals contaminated with the poison showing signs of stress marked by unstable movement and a tendency to be at the bottom[55,56].

According to[57,58], the death of test animals does not always come from one factor alone but a synergistic phenomenon, where there is a combination of two or more substances that strengthens the toxicity. The response that occurs when xenobiotic substances interfere with cell / sub-cell processes in living things to the extent that causes instant death is a lethal effect[59,60].

The smallest value of LC50-96h in D. magna for diazinon-formaldehyde-IPA is 11.82, in accordance with the biodegradability of diazinon-formaldehyde-IPA which is included in the non-biodegradable category. Sucrose-glycerol-acetic acid with the largest LC50-96h value is 567.88 according to the biodegradability of sucrose-glycerol-acetic acid which is included in the biodegradable category. The LC50-96h toxicity test has verified the results of laboratory analysis.

4. Conclusion

The correlation between Pow and BOD/COD ratio was found to be negatively correlated in the range of 0.1 - 0.9 for 6 types of mixtures of organic substances. A mixture of organic substances that are more biodegradable will tend to dissolve in water. Diazinon-formaldehyde-IPA had the lowest BOD / COD ratio (<0.2) and the highest Pow value (>4). This is also supported by the results of the LC_{50}-96h toxicity test against *D. magna*, with the smallest LC-50 value of 11.82 mg/L. Meanwhile, the mixture of sucrose-glycerol-acetic acid organic substances had the highest BOD/COD ratio (> 0.7) and the lowest Pow value (<0.7) with the highest LC-50 value of 567.88 mg/L. The mixture of other organic substances had a characteristic value between sucrose-glycerol-acetic acid and diazinon-formaldehyde-IPA. The Pow variability and the BOD/COD ratio had a negative correlation. A mixture of organic matter was more biodegradable and had a higher tendency to dissolve in water.

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