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Selita Pasaribu
Jl. Raya Bandung Sumedang, KM 21, Hegarmanah, Jatinangor, Sumedang, Faculty of Fisheries and Marine Science, Universitas Padjadjran West Java, Indonesia

Zahidah Hasan
Jl. Raya Bandung Sumedang, KM 21, Hegarmanah, Jatinangor, Sumedang, Faculty of Fisheries and Marine Science, Universitas Padjadjran West Java, Indonesia

Yuniarti
Jl. Raya Bandung Sumedang, KM 21, Hegarmanah, Jatinangor, Sumedang, Faculty of Fisheries and Marine Science, Universitas Padjadjran West Java, Indonesia

Heti Herawati
Jl. Raya Bandung Sumedang, KM 21, Hegarmanah, Jatinangor, Sumedang, Faculty of Fisheries and Marine Science, Universitas Padjadjran West Java, Indonesia

Corresponding Author:
Selita Pasaribu
Jl. Raya Bandung Sumedang, KM 21, Hegarmanah, Jatinangor, Sumedang, Faculty of Fisheries and Marine Science, Universitas Padjadjran West Java, Indonesia

The effectivity rough horsetail (*Equisetum hymale*) and Mexican sword (*Echinodorus paleafolius*) as phytoremediation agent in reducing heavy lead metal (pb) in the upper Citarum River – Daeyeuhkolot

Selita Pasaribu, Zahidah Hasan, Yuniarti and Heti Herawati

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Abstract
The purpose of this study was to find the most effective between rough horsetail (*Equisetum hyemale*) and Mexican sword (*Echinodorus paleafolius*) as a phytoremediation agent in reducing heavy metal lead (Pb). The method used in the study was an experimental method with a completely randomized design (CRD) consisting of four treatments and three replications using two aquatic plants, namely rough horsetail and Mexican sword. The physical and chemical parameters observed were temperature, degree of acidity (pH), dissolved oxygen (DO), and the concentration of heavy metal lead (Pb). The optimal research results on the combination treatment to reduce the concentration of heavy metal lead (Pb) was 0,0066 mg/L with a decrease proportion of 90,32% for 14 days. The best phytoremediation results in the rough horsetail and combination treatment used for fish with fish survival was 100%.

Keywords: Citarum River, Heavy Metal Lead (Pb), Mexican sword, phytoremediation, rough horsetail

1. Introduction
Rivers are an important component of human life. However, there are developments in various fields such as industry, agriculture, fisheries, and community activities that do not pay attention to the environmental impacts that result in river pollution. One of the biggest contributors to the burden of pollutants to the river is the wastewater industry. Based on the BBWS-Citarum in 2014 there are 64 industries around the Citarum River Dayeuhkolot segment, namely the textile industry that produces solid and liquid waste, such as electronic device factories. Also, the statement of [1] the percentage of wastewater industry disposal to the Citarum River reached 48% with the average waste disposal has exceeded the quality standards set.

Citarum River contains lead heavy metals (Pb) with the highest concentration found in Citrarum River Dayeuhkolot Segment which is 0,024 mg/L [2]. Citarum River pollution is increasing for longer if there is no improvement. Phytoremediation is an effort that can be made for the restoration of the environment by using a plant and collaboration with microorganisms that will change harmful substances into harmless to the environment [3]. Especially for aquatic animal habitats. Because excess heavy metals are highly toxic to animals and plants and may be associated with adverse health effects [4]. Carp is the animals that carp are sensitive to environmental changes, and carp is a fish that carp is susceptible to heavy metal [5]. One of the plants that can be used is rough horsetail that is effectively used as a heavy metal-binding agent such as Pb and Cr [6]. Besides that, the mexican sword can be used as a phytoremediation agent. Because roots on mexican sword have the advantage of being able to absorb heavy metals [7].

The purpose of this research was to find the most effective plants between rough horsetail and mexican sword as a phytoremediation agent in reducing heavy metal lead (Pb).

2. Materials and Methods

2.1 Time and Location

The research was conducted from October to December 2020. The research was conducted at Ciparanje Greenhouse, Universitas Padjadjaran. The water sample used from citarum river water upstream-Dayeuhkolot.

2.2 Research Method

The research method used in this research was the experimental method with study used a completely randomized design (CRD) with 4 treatments and 3 repetitions. Materials (rough horsetail, mexican sword, carp, citarum river water, aquadest, coral) and tools (aquarium, *fiber* tub, aerator, dissolved oxygen meter, pH meter, thermometer, AAS (*Atomic Absorpstion Spektofotometer*), analitycal scale, ruler, jerigen, measuring glass.

2.3 Data Analysis

The water quality parameters measured include chemical-physical water parameters i.e temperature, pH, and dissolved oxygen are measured directly (*in situ*). While the analysis of the content of lead-heavy metals (Pb) in citarum river water is indirectly (*ex-situ)*.

Calculation of the percentage of lead-heavy metal (Pb) removal according to [8] as follows:

$$\% \text{ removal} = \frac{\text{initial metal concentration} - \text{final metal concentraction}}{\text{initial metal concentration}} \times 100\%$$

The percentage of rough horsetail and mexican sword growth during a specific growth rate is calculated using the formula [9]:

$$\% \text{ Wt/day} = \frac{\text{Final wet weight} - \text{Initial wet weight}}{\text{Initial wet weight x t}} \text{ x } 100\%$$

Survival fish in the research after the phytoremediation are using the survival rate (SR) [10]:

$$SR = \frac{\text{Number of final survival rate}}{\text{Number of initial survival rate}} x\ 100\%$$

Data analysis was done by a descriptor. Analysis of lead-heavy metals (Pb) is conducted by the AAS method. The recommended quality standard of Indonesian Government Regulation No. 82 of 2001 class II and III is used as a comparison with the results of water sample data analysis. One-way ANOVA was used to analyze the results of the decrease in the concentration of timbal heavy metals (Pb). It was done by using Duncan's honestly significant difference test at $p < 0{,}05$.

3. Results and Discussion

The government is making efforts to overcome the decline in the water quality of citarum river. The program that was proclaimed by Indonesian Government Regulation No. 15 of 2018 concerning the acceleration of pollution and damage control in the Citarum River area, with the program name "*Citarum Harum*" [11]. "Citarum Harum" program implemented in the form of land revitalization and things related to tributaries. The Citarum Harum program is carried out by central and regional agencies involving the Indonesian National Army [12]. Besides efforts made by the government, water quality improvement can be done by phytoremediation activities using water plants. So that this phytoremediation activity can be used as an additional source of information about improving the water quality of the Citarum River [13].

3.1 Water Quality Parameter

Phytoremediation was conducted during 14 days at Greenhouse Ciparanje Universitas Padjadjaran.

3.1.1 Temperature

The result of temperature measurements before phytoremediation using plants was 28ºC. The statement of [14] temperatures range from 28-29ºC optimal for the development of organisms such as fish in tropical waters.

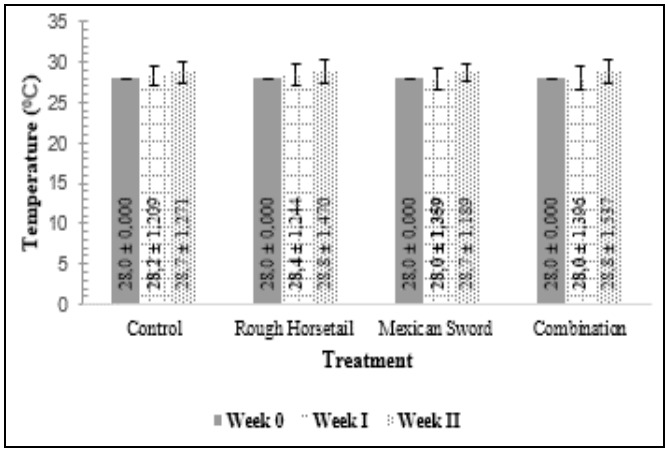

Fig 1: Temperature Value

The results of phytoremediation observations do not see a decrease or increase in temperature fluctuating between treatments (Fig 1). The average water temperature in the control treatment in the first week was 28,2±1,209ºC and experienced a temperature in the second week of 28,4±1,271ºC. The average water temperature in rough horsetail treatment in the first week was 28,4±1,244ºC and increased in the second week by 28,8±1,470ºC. The statement of [15], the optimum temperature range for the growth of rough horsetail is 28-36 ºC because the temperature conditions are buried optimum for the process of biodegradation in rough horsetail. The average water temperature with mexican sword treatment in the first week was 28,0±1,359 ºC and increased in the second week by 28,7±1,189 ºC. The statement of [16] the optimum temperature range for the growth of mexican sword at a temperature of 25-35 ºC. The average water temperature in the combined treatment of rough horsetail treatment and mexican sword in the first week was 28.0±1.396 ºC and the second week was 28.8±1.537 ºC. The Temperature value is appropriate standards of quality class II and III by the Indonesian Government Regulation No. 82 of 2001, which is the temperature used in fisheries ranged from 3 deviations.

3.1.2 Acidity (pH)

The pH value of water can be used as one of the limiting factors because each organism has a maximum tolerance and minimum pH value of the water so that it is used to analyze the good or bad of water [17].

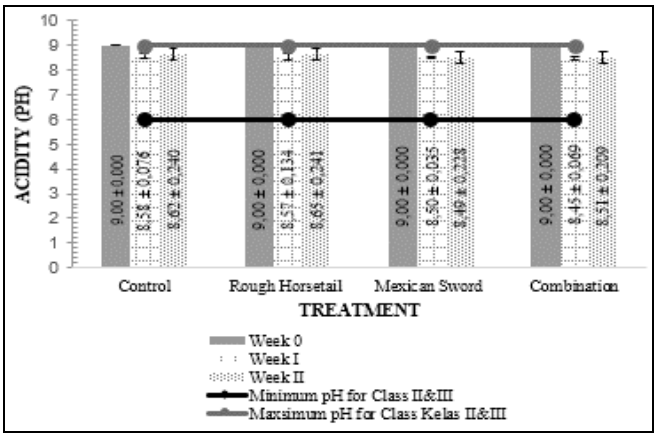

Fig 2: Value of pH

The pH value measured before phytoremediation is 9, which indicates that it can conduct fishery activities in these waters but is less than optimal based on Indonesian Government Regulation No. 82 of 2001 with pH values for class II and III for fisheries activities ranged from 6-9. The average weekly pH value for each treatment is shown in Fig 2. The results of the first week of research, the pH value decreased, with a low pH in the combination treatment of 8,45 ± 0,069 and the highest in the control treatment was 8,58 ± 0,076. While observations by the second week, the average pH value increases, with an average pH of the highest in the treatment of rough horsetail 8.65 ± 0.241 and the lowest in Mexico sword treatment of 8,49 ± 0,228. However, the changes in pH values did not experience excessive fluctuations in observations of the first week and the second week. Conditions range pH 8-8,5 can usable for the growth of rough horsetail water and mexican sword and support microorganisms for decomposition of organic matter in the waters. According to [18] pH value, 6-8 is the optimum pH for rough horsetail growth because the plants absorb the elements in the planting medium. While according to [16] pH 4,5 8 is the pH value for optimum mexican sword growth.

3.1.3 Dissolved Oxygen (DO)

Dissolved oxygen (DO) was an important parameter for aquatic organisms. According to [19] if dissolved oxygen decreases in the waters will be dangerous especially for aquatic life.

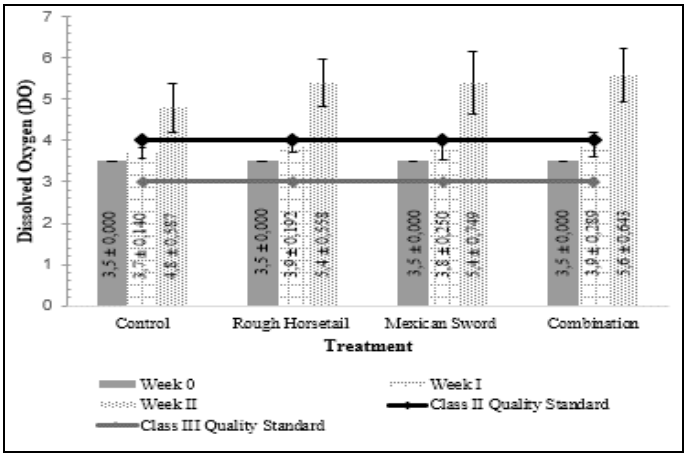

Fig 3: Dissolved Oxygen Concentration

The value of dissolved oxygen (DO) before phytoremediation is 3,5 mg /L (Figure 3), the initial DO in accordance with class II quality standards but not yet in accordance with class III quality standards based on Indonesian Government Regulation No. 82 of 2001 i.e 4 mg/L and 3 mg/L. So that the condition of the water before the phytoremediation can not be used as a place of fishery activities. Dissolved oxygen value (DO) when phytoremediation in the first week with control treatment of 3,7±0,140 mg/L, rough horsetail treatment of 3,9±0,192 mg/L, mexican sword of 3,8±0,250 mg/L, and combination treatment was the highest average DO value in the first week of observation of 3,9±0,289 mg/L. The average DO in the second week of observation increased in each treatment with an average DO control treatment of 4,8±0,558 mg/L, rough horsetail treatment of 5,4±0,749 mg/L, mexican sword of 5,4±0,749 mg/L, and a combination treatment of 5,6±0,643 mg/L. The results in the second week have met the quality standards of class II and Class III according to Indonesian Government Regulation No. 82 of 2001 so that water samples can be used for cultivation activities.

3.1.4 The concentration of Lead Heavy Metal (Pb)

The concentration of heavy metals lead citarum river water before the phytoremediation of 0,0682 mg/L, this result is above the quality standard of class II and III in Indonesian Government Regulation No. 82 of 2001 for fishery activities that are 0,03 mg/L.

Table 1: The second week of the Pb concentration statistical test

Treatment	Concentration Pb	Notation
Control	0,0546	a
Mexican Sword	0,0146	b
Rough Horsetail	0,0093	c
Combination	0,0066	c

Description: Heavy metal concentrations of lead (Pb) by Duncan's Test

The statistical results of the second week of lead-heavy metal (Pb) concentrations are significant differences between treatments. So it continued with Duncan's honestly significant difference test at $p < 0,05$ (Table 1). The results showed that the control treatment was significantly different from the mexican swords treatment, rough horsetails, and combinations. The treatment of the mexican sword differs significantly from the treatment of the rough horsetail and mexican sword, while the rough horsetail does not differ significantly from the combination.

The results of heavy metal concentration after the phytoremediation process in the first and second week decreased (Fig 4). The highest concentration of heavy metals in the control treatment was 0,0653±0,001 mg/L with a percentage decrease of 4,30% in the first week and $0,0546±0,003^{a}$ mg/L with a percentage decrease of 19,94% in the second week. The result of the control treatment still does not meet the quality standards of Indonesian Government Regulation No. 82 of 2001 in class II and III for fishery activities. While the lowest concentration of lead-heavy metal is with the combined treatment of rough horsetail and mexican sword which is 0,0135±0,002 mg/L with a percentage decrease of 80,16% in the first week and $0,0066±0,001^{c}$ mg/L with a percentage decrease of 90,32% in the second week.

The result of the concentration of lead-heavy metal with the addition of rough horsetail and mexican sword treatment has met the quality standards of class II and III in Indonesian Government Regulation No. 82 of 2001 so that rough

horsetail and mexican sword work optimally in lowering the concentration of lead-heavy metal. According to [20] rough horsetail can lower the concentration of lead metal (Pb) to 82,2% (60 plant stems with a batch system). While the percentage decrease in heavy metals chromium (Cr) is 61,2% (60 stems of plants with a continuous system). In line with the research [13] rough horsetail plants at the age of 30 days can set aside the content of lead-heavy metals (Pb) by 76%. According to [7] the percentage decrease in lead heavy metals (Pb) using mexican sword reached 81,72%.

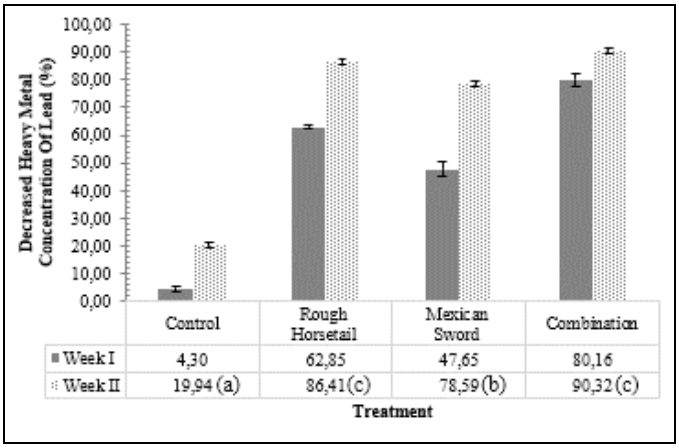

Fig 4: Percentage Decrease of Lead Heavy Metal Concentration (Pb)

Water quality parameters, namely dissolved oxygen (DO), pH, and temperature in the observations, do not differ much between treatments, meaning that water quality is not affected by the absorption of lead-heavy metals (Pb) in each treatment. However, the effect on the absorption of heavy metal concentrations in the plant as a phytoremediation agent. According to [21], the ability of plants to lower lead heavy metals in water is influence by the morphological structure of stems and plant roots. According to [20] the rough horsetail has the advantage of a stem containing silicate, which effectively binds heavy metals from wastewater. While the advance of mexican sword plants is that water jasmine can stabilize heavy metals performed on the root system [22]. So as the phytoremediation process with the treatment of a combination of rough horsetail and mexican sword is more effective to lower the heavy metal lead.

3.1.5 Plant Growth

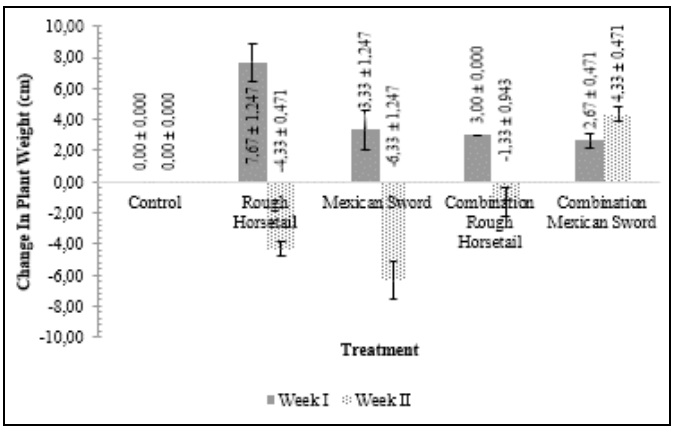

Fig 5: Changes in Plant Weight

The results of measuring plant weight in the first week experienced weight in each treatment (Fig 5), with the highest weight in the rough horsetail treatment, namely 7.67 ± 1.247 grams. The lowest weight in the treatment of the mexican sword combination was 2.67 ± 0.577 grams. However, in the second week, there was a decrease in the fresh weight, with the decrease of rough horsetail treatment by -4,33±0,471 grams, mexican sword by -6,33±1,247 grams, and rough horsetail combination treatment by -1,33±0,943. The shrinkage of the weight in the second week on the rough horsetail and mexican sword treatment is suspected stems and leaves that begin to dry and turn yellow due to too long exposure to the burden of pollutants. According to [22], the ability of plants in the phytoremediation process in the absorption of heavy metals can inhibit plant growth and cause physiological symptoms such as dry leaves. It also was supported research by [23] hat excessive exposure to heavy metals in plants resulted in increased concentration of heavy metals in plants and decreased plant quality. The combined treatment of the mexican sword in the second week increased by 4,33±0,471 grams, indicated by the increase in plant weight of the second week of plants on the combined treatment of the mexican sword characterized by the growth of new shoots on the mexican sword.

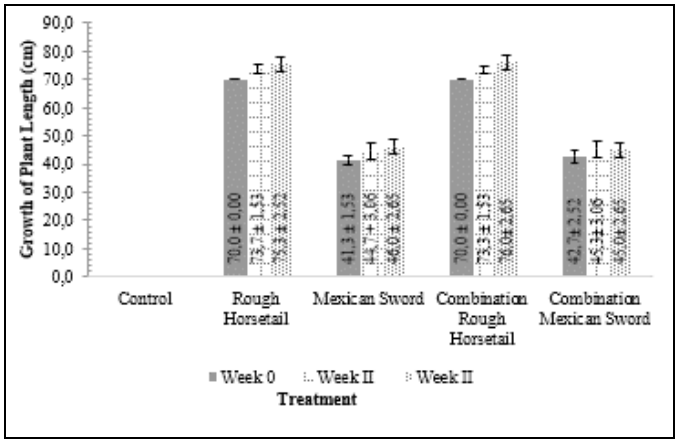

Fig 6: Growth of Plant Length

The length of the plant is observed for 14 days and measuring once every seven days (Fig 6). During the research, the average length of the plant increased. The highest length growth in the first week was the treatment of rough horsetail at the beginning of observation 70,0±0,00 cm to 73,7±1,53 cm. Meanwhile, the lowest length growth in the first week was the combination treatment of the mexican sword at the beginning of observation 42,7±2,52 cm to 45,3±3,06 cm. The measurement results from the second week of the highest growth with a rough horsetail combination treatment of the water that is 70.0±0.00 cm become 76,0±2,65 cm. Meanwhile, the lowest long growth was the combination treatment of the mexican sword at the beginning of observation 42,7±2,52 cm to 45,0±2,65 cm.

3.1.6 The Survival of The Carp

Carp used as test fish in water phytoremediation process results with the fish size is 3-5 cm. Carp that are less than three months old are very sensitive to environmental changes because at the time of fish seed can respond to toxic material pollution [5]. Carp are fish that are sensitive to changes in dissolved oxygen [24]. Low dissolved oxygen concentration can lead to increased toxicity of ammonia and the impact on fish growth slowing [25].

Carp used for the test is ten heads for each treatment and

maintained for seven days (Figure 7). The results of phytoremediation with a combination and rough horsetail had the highest survival rate (SR) of 100%. While the second highest SR value was in the mexican sword treatment of 97%, and the lowest SR was in the control treatment of 77%. Therefore, the survival of the best of the four treatments, namely phytoremediation with rough horsetail treatment and combination treatment. According to [26], the survival of carp in phytoremediation using water lettuce ranges from 52-88%.

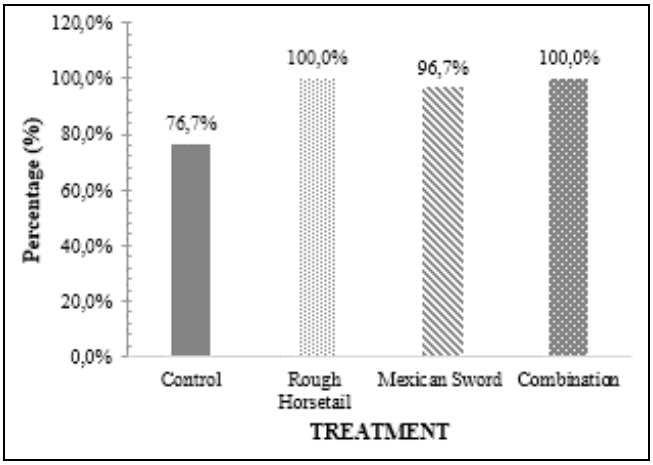

Fig 7: Survival Rate of The Carp

4. Conclusions

Phytoremediation with a combination treatment of rough horsetail and mexican sword is the most effective treatment to lower the concentration of lead-heavy metals, with a concentration of lead-heavy metals 0,0066 mg/L with a percentage decrease of 90,32%. The concentration of lead-heavy metal in the control treatment became 0,0546 mg/L with a decreasing percentage of 19,94%. The rough horsetail treatment was 0,0093 mg/L with a percentage decrease of 86,41%, treatment with mexican sword was 0,0146 mg/L with a percentage decrease of 78,59%.

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