Preliminary Design of Phytoremediation of Phosphate From Liquid Fertilizer Waste by Duckweed (Spirodera sp.) and Yellow Iris (Iris pseudacorus L.) Integrated with Processing System to Produce Value Added Bioproducts Using Biorefinery Concept

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Abstract—Agriculture is one of the biggest economy sectors of Indonesia, with total area used for agriculture reaches 8.19 million hectares. Usage of fertilizer in agriculture causes contamination in the environment, with one of the biggest contaminants takes form as excess phosphate. This happens because phosphate is one of the most intensely used fertilizer and also has the lowest absorption rate by plant. Excess phosphate can cause eutrophication on bigger body of water. One method to reduce this effect is by doing phytoremediation, with *Spirodela* sp. and *Iris pseudacorus* L. This method able to reduce phosphate concentration from 29.625 mg/l to 0.2 mg/l. By applying biorefinery concept, plants biomass will be used; *Spirodela* sp. would be used to produce duckweed powder with yield of 20.8%, while *Iris* plant will be extracted its flavonoid content to produce flavonoid powder, with yield of 20.9%. These byproducts add the economic value of the system, with GPM of 5.91. This shows that applying biorefinery concept to phytoremediation activity is profitable.

Keywords—phytoremediation; biorefinery; excess phosphate; value-added byproducts

I. INTRODUCTION

Agriculture is one of the most prominent economic sector in Indonesia. According to the publication released by Agricultural Ministry of Indonesia, land area used for agricultural practices reach 8.19 million hectares in 2017. This intense activity also requires a great amount of fertilizer, which is used as plant nutrient source. Although required, fertilizer often used excessively, causing nutrition left unabsorbed by plant left in the soil. These leftovers can contaminate water and soil, and by rain or water run off the contamination can spread to bigger body of water. One of the intensely used fertilizer is the one that contains phosphate, since it has great use in plants, however, it is relatively low absorbed by plants, reaching only 15% of total phosphate in fertilizer can be used. Phosphate left unabsorbed usually taken away by water runoff and causing eutrophication in lake and river [1].

One of removal methods for this contaminant is by phytoremediation. It is a technology that used plants to reduce or diminish the effect of contaminants in the water and soil. In the process, factors that determine the feasibility of this method is contaminants property, level of contamination, contaminated area condition, and plants used as phytoremediator [2]. *Iris pseudacorus* L. is an acaulescent plant, has long leaves, and has rhizome root, with height reaches 1 meter. This plant typically lives in wetland, and other shallowly submerged land. A research shows that its ability to phosphate remediation ranges from 25%-34.17% [3]. This shows that *Iris pseudacorus* L. is potential to be used as a phytoremediator. Duckweed (*Spirodela* sp.) is a plant with a simpler morphology, and is consisted by a few oval shaped, approximately 5 mm in size leaves. This plant lives in colony and has short growing time. Duckweed also potential to be used as phytoremediator. One research shows that duckweed reduce TSS, BOD, COD, excess nitrate, ammonia, Cu, Pb, Cd, and Zn consecutively: 96.3%, 90.6%, 89.0%, 100%, 82.0%, 64.4%, 100%, 100%, 93.6% and 66.7% [4].

Constructed wetland is one application of phytoremediation concept that uses wetland plants or other aquatic ones as its phytoremediating agent. In this research, sub surface flow (SSF) type of constructed wetland is used. This system uses submerged plants as its filter. Plants used as phytoremediator can be further processed to reduce waste by utilizing biorefinery concept. This concept uses a transformation of a biological resources to other products while increasing its economic value [5]. In its application, duckweed can be
used as high protein powder, while Iris plant can be extracted its flavonoid content. Flavonoid is useful as anti-oxidant and anti-cancer substances. Aside from reducing phosphate contaminant, a phosphate phytoremediation system that is integrated with biorefinery concept can increase economic value of plants that are used as phytoremediator.

II. MATERIALS AND METHODS

A. Analysis of Phosphate Removal Efficiency using Phytoremediation

This research analyzed emerging potential of phytoremediating excess phosphate using duckweed, *Spirodela* sp. and Iris plant, *Iris pseudacorus* L. Duckweed was retrieved from Azola Purwodadi while Iris plant was obtained from School of Life Sciences and Technology, Institut Teknologi Bandung. This research was conducted at Instructional Laboratory of Bioengineering Department, Institut Teknologi Bandung.

Phytoremediation was done in the time span of 2 weeks, with *Spirodela* sp. as its phytoremediator for the first week, and *Iris pseudacorus* L. for the next. Growth was measured from dry weight and fresh weight. Phosphate concentration was measured in the growth medium and the biomass of phytoremediating agent with method developed by Kuttner & Cohen [6].

B. Design of Process Flow Diagram

Process flow diagram is a diagram to illustrate production process and its production units. This research explains excess phosphate phytoremediation that is integrated with protein supplement production and flavonoid extract. Process Flow Diagram in this research was made by SuperPro Designer® software.

C. Mass Balance Analysis

This analysis is used to estimate inflow of substrate and outflow of products in the process. In this research, mass balance analysis estimated the amount of plant biomass result from phytoremediation activity and their byproducts, which are protein supplement and flavonoid extracts.

D. Economic Analysis

Economic analysis is used to examine the feasibility of this biorefinery based production system. Analysis method used in this section is by calculating gross profit margin (GPM). This number was calculated from the ratio of increment of product and raw material price divided by raw material price as shown in Eq. (1).

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GPM = \frac{(product\ price - raw\ material\ price)}{raw\ material\ price}
\]  

(1)

III. RESULTS AND DISCUSSION

A. Analysis of Phosphate Removal Efficiency using Phytoremediation

Growth of phytoremediator and phosphate concentration in growth medium and biomass of phytoremediator was measured every three days in the time span of two weeks. Below is the data of dry and fresh weight for *Spirodela* sp.

![Growth of Spirodela sp. according to its fresh weight (a) and dry weight (b).](image)

From the data above, it can be derived the specific growth rate for *Spirodela* sp. was 1.1329 gram day⁻¹. There was no sign of deceleration of growth of *Spirodela* sp., therefore it can be concluded that growing *Spirodela* sp. in excessive phosphate condition does not give inhibitory effect.
Growth of *Iris pseudacorus* L. was investigated in the same fashion. From the data, it is shown that growing *I. pseudacorus* in excess phosphate medium gave specific growth rate of 3.6832 gram day$^{-1}$. Thus growing *I. pseudacorus* in an excess phosphate condition does not give inhibitory effect.

Phosphate concentration in growth medium was assessed with spectrophotometry method using ammonium molybdate and SnCl$_2$ reagent. Addition of these reagents would result in reaction of phosphate ion and creating ammonium phosphomolybdate complex. This complex then would be reduced by SnCl$_2$, resulting a distinguishable shade of blue from molybdenum. This molybdenum would be the indicator of the amount of phosphate in the mixture and assessed by spectrophotometry to obtain its absorbance. By creating a standard curve, the concentration in the mixture can be known [7]. Data for the absorbance in this assay are presented in Fig. 3.

The constructed wetland system design that is used is horizontal subsurface flow (HSF) system. This constructed wetland system designed with several important parameters, such as the depth of wetlands, the wetland area, and hydraulic residence time. The depth of HSF wetland has been taken as 40 cm and the porosity of the substrate as 40%. Using the basis of average daily influent flow rate of wastewater (Q) 40 m$^3$/day, the constructed wetland sizes can be determined using Eq. (2) [8].

$$A_h = \frac{Q_d \ln(C_e - \ln C_i)}{K}$$  

(2)

where $A_h$ is the surface area of wetland (m$^2$), $Q_d$ is the average daily flow rate of wastewater (m$^3$/day), $C_i$ is the influent phosphate concentration (mg/l), $C_e$ is effluent phosphate concentration (mg/l), and K is first order rate constant (m/day). It is assumed that the average daily influent phosphate concentration ($C_i$) is 29,625 mg/l [9], and we wanted to reduce the effluent phosphate concentration ($C_e$) up to 0.2 mg/l by using phytoremediation method. K can be determined using Eq. (3) and (4) stated below:

$$K = K_T d n$$  

(3)

$$K_T = K_{T0} (1.06)^{\frac{T - 20}{20}}$$  

(4)

with $K_{T0}$ is first order rate constant at 20ºC (day$^{-1}$) = 1.1 day$^{-1}$, $T$ is the operational temperature of system (25ºC), $d$ is the depth of water column (m), and $n$ is the porosity of the substrate medium (expressed as fraction). Given that the average value of $d$ is 0.4 m and $n$ is 40% or 0.4, as recommended by UN-Habitat [10], the value of $K_T$ can be determined which is 0.236 m/day. Thus, the
value of $K_T$ could be used to find the value of the surface area of wetland ($A_h$) which is 847.128 m$^2$.

Stagnant pool in a wetland can be caused by poor flow pattern. This condition can lead to accumulation of scum and mosquito breeding. A solution to this is by designing multiple flow paths, so that the system is broken down to units that are easier to maintain. In general, an optimum length to width ratio of a constructed wetland is 5:1 [11]. A geometrical design of constructed wetland is shown in Fig. 5.

![Fig. 5. Design criteria of the constructed wetland system.](image)

Hydraulic residence time is the average time that the wastewater influent remains in the wetland system, and it is correlated directly to the effectivity of contaminant’s removal in the wetland system [12, 13]. Hydraulic residence time ($t$) can be determined using Eq. (5).

$$ t = \frac{L \cdot w}{A_h} $$  \hspace{1cm} (5)

where $L$ is the length of wetland system that is parallel to flow direction (m) = 65.08 m and $w$ is the width of wetland system (m) = 13.016 m. Given that the value of $L$ is 65.08 m and $w$ is 13.016 m, the value of hydraulic residence time ($t$) can be calculated that is 3.389 days for each path.

B. Process Flow Diagram

Fig. 6 shows the diagram of process. Duckweed biomass will be washed and dewatered to reduce contamination possibility, then dried in a certain temperature to obtain the dry form [14]. Then, dried duckweed will be grinded and milled to certain size, resulting in powdered form of duckweed. This product can be used in other food industry as high protein supplement. As for *I. pseudacorus* after washed and dewatered, it will be dried in room temperature (23–34°C) then grinded to powder. This powder then will be extracted its flavonoid content by submerging it in 75% ethanol for 24 hours. Solid waste of this extraction will be separated from ethanol and used as organic compost. Liquid extract of *I. pseudacorus* will be freeze dried and powdered then stored in -4°C for preservation [15].

C. Mass Balance Analysis

In this mass balance analysis, we used the area of 1 yellow iris 40 cm x 40 cm, weight of 1 yellow iris 100 gram [16], and density of duckweed 1kg/m$^3$ as basis of calculation. About 40000 liters of wastewater will be flown through phytoremediation system 1. With this, duckweed will grow into about 423 kg, assuming the growth rate of duckweed to be similar. Duckweed biomass then will be washed and dewatered and dried then powdered, resulting in 88.27 kg of dry duckweed powder. Wastewater from system 1 will be processed again with system 2, containing *Iris pseudacorus* L. Iris will grow into about 275 kg. This biomass then will be washed and dried then extracted its flavonoid content with the help of 423 liters of methanol 75%, resulting in 170.5 kg flavonoid extract. Solid waste from this extraction is used as organic compost, with the amount of 123.2 kg. Flavonoid extract will be dried and added maltodextrin, resulting in the powder form of extract with the amount of 57.6 kg.

![Fig. 6. Process flow diagram.](image)
This production results in a GPM of 5.919, showing that this activity is deemed as profitable.

IV. CONCLUSIONS

In this research, developing an excess phosphate phytoremediating system with *Spirodea* sp. and *Iris pseudacorus* L. that is integrated with biorefinery concept would reduce 29.625 mg/l phosphate to 0.2 mg/l in 40000 liters of work volume, resulting in water with acceptable quality according to Indonesian standard. Aside from that, this system also produces 88.27 kg duckweed powder that can be used as protein supplement, and 57.6 kg flavonoid powder extract from *Iris pseudacorus* L., also 123.2 kg organic compost per batch. These products are counted as value-added byproducts that improve the economic value of this system, with 5.92 GPM. This research shows that application of biorefinery concept is not only able to reduce the negative impact of excess phosphate waste, but also provides economic value, thus reinforcing the sustainability of this system. One remarkable note that this research shows that it is possible to do environment remediating activity while also getting economic benefit, contradicting the popular beliefs that such practices are costly and unprofitable. It is encouraged for not only us but also other prospecting parties to do a more extensive research and application of this topic as this research is only a preliminary view of what the system would be.

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