Effectivity of water hyacinth (*Eichhornia crassipes* (Mart.) *Solms*) as bio filter for microbiological water quality in Purwomartani Housing, Kalasan, Sleman, DIY

Y V Kahol, N S Ningrum, E Retnaningrum, S Darmasiwi*

Laboratory of Microbiology, Faculty of Biology, Universitas Gadjah Mada, Yogyakarta, Indonesia

Email: saridarma@ugm.ac.id

**Abstract.** Water is one of the most important needs for organisms. However, microbial contamination in water has become serious issue recently. We were interested to reduce the microbial contamination in water by using biological agents such as water hyacinth. In some studies, water hyacinth has antimicrobial activities as well as their function as phytoremediation agent. This research was done to study the effectiveness of water hyacinth (*Eichhornia crassipes* (Mart.) Solms) as bio filter against coliform in water of Mataram water channel in Yogyakarta. The bio filter were arranged as following layer; gravel, zeolite and water hyacinth as the most upper part of bio filter. It was then set with the direction along with the river flow. The water samples were then taken triplicates, each from the upstream (before bio filter treatment) and downstream (after bio filter treatment). It were then continued by Most Probable Number (MPN) test which include the presumptive, confirmed, and completed test. The MPN results showed that coliform in the upstream were found about 57.34 ± 54.35 MPN / 100 ml while in the downstream were about 808 ± 678.96 MPN / 100 ml. The statistical test showed that the difference between upstream and downstream were not significant. Thus it can be concluded that water hyacinth bio filter was not effectively used as bio filter against coliform bacteria.

1. Introduction

The life cycle depend on the availability of water because most of organisms need water to perform their chemical reaction. However, this essential role of water has been challenged serious problems. The rising of population and growth of civilization has made demand of water increasing vastly [1]. Moreover, availability of clean water become hard to be met due to the environmental pollution and contamination as side effect of human activities. Thus, it should be our concern to protect water from more dangerous effect and scarcity.

In small scale i.e. household, water is often polluted by microorganisms in consequence of daily waste production. Contamination of water-borne pathogen has been concerned worldwide since it is major cause of water-borne diseases such as diarrhea, cholera, gastrointestinal illness. The water-borne diseases are mostly correlated by the existence of indicator and pathogenic bacteria, for example *Cryptosporidium, Giardia lambia, Legionella*, Total coliform and other strain of pathogenic bacteria. This amount and type of bacteria contained in water varies depending on water source and its environment [2].
As one of the growing city in Indonesia, Yogyakarta has now become one of destination for urbanization. One of the landmark and populated area in Yogyakarta is Mataram Canal, which also crossing the Purwomartani Housing, Kalasan, Sleman, Yogyakarta. Formerly this area was an agricultural area, and the canal function was for irrigation, but due to the urban growth currently water in these canal are expected to be polluted by daily human activities [3].

To overcome water pollution and contamination problem, there were many research study about the application of adsorption, magnetic assistance, membrane, plant coagulation and plant vegetation treatment methods to purify the water [4,5,6,7,8,9]. The use of physical and mechanical method for water purifying probably quite effective to drawback the water contamination problem but apparently it is not cost effective and require high technology for developing countries.

Biofilters, also known as bioretention systems or raingardens, are soil-plant based systems that promote infiltration and evapotranspiration [10] In case to develop this technology, plants from species of *Juncus effusus* L., *Scirpus validus* L., and *Typha latifolia* L. were reported to be effectively used for wastewater treatment in wetlands area. These plants has been known for their potential to remove turbidity and their ability as disinfectants in water [9]. Similarly, we would like to study about the use and effectivity of water hyacinth (*Eichhornia crassipes* (Mart.) Solms) as bio filter for purifying water along Mataram Canal, specifically in Purwomartani Housing, Yogyakarta.

Water hyacinth is a plant that lives on the surface of the water. This plant is generally found in tropical regions including Indonesia. Water hyacinth are popular as phytoremediation agent and their antimicrobial effect [11,12]. As far as our knowledge, there had never been any research on water hyacinth as a microbial bio filter in water before. Therefore, this research aimed to determine the effectiveness of water hyacinth (*Eichhornia crassipes* (Mart.) Solms) as a bio filter on microbiological quality in residential waterways in Purwomartani area, Kalasan, Sleman, Yogyakarta.

2. Materials and Method

2.1. Bio filter design
Bio filter was designed as following layer: gravel, zeolite and water hyacinth as the most upper part of bio filter. It was then set with the direction along with the river flow.

![Figure 1. Water Hyacinth Bio filter Design.](image)

2.2. Water sampling
Water sampling and bio filter treatment were done in sub-area of Mataram Canal in Purwomartani Housing, Sleman, Yogyakarta, along upstream segments (-7.7689132, 110.4572752) to downstream segments (-7.7651221, 110.4695456) coordinates. Water samples were taken at 2 (two) points, before the bio filter treatment (upstream) and after the bio filter treatment (downstream). Each point were taken triplicate. The water samples were taken using 100 ml dark bottle. Samples were then taken to the Laboratory of Microbiology, Faculty of Biology UGM for the analysis.
2.3. Bacteriological examination
The bacteriological examination were performed by using MPN methods [13]. This method were consisted of presumptive test, confirmed test and complete test.

2.3.1. Presumptive test. Each water sample were diluted in serials $10^{-3}$ mL, $10^{-4}$ mL, $10^{-5}$ mL in Lactose Broth medium. Samples were incubated for 48 hours at 30°C. Positive tubes of coliform were shown by medium change from red into yellow color. Positive samples were analyzed further into confirmation tests.

2.3.2. Confirmed Test. Positive samples from presumptive tests were inoculated using streak plate method into Endo Agar medium. Samples were incubated at 24 hours at 35°C. Positive results of coliform were shown by golden or green metallic colonies. The positive colonies were inoculated into nutrient agar medium to be observed further to complete test.

2.3.3. Complete Test. Positive samples from confirmed test were then analysed using Gram staining test and continued by microscopy observation.

2.3.4. Growth Characterization. Microbial growth were observed using Nutrient Agar medium. The observation include colony shapes, elevation, colony edge, internal structure, and cell morphology.

2.4. Data Analysis
Data of number of positive tube series in the MPN test were compared with MPN table to estimate amount of coliform. Data were analysed using one way ANOVA continued by Duncan test in order to test the significance value.

3. Results and Discussion
In order to know the level of water quality and to measure the effectivity of water hyacinth as bio filter for water quality, we used MPN (Most Probable Number) test which quantify the level of coliform in water. As we know that coliform was chosen as biological indicator for water quality because of their presence can be found universally from human to other mammals in extensive numbers, easy to detect in low cost, and also the bacteria will not multiply outside the hosts body [14].

We compared the results of MPN, before and after treatment of water hyacinth bio filter. The results can be shown below (Table 1).

**Table 1.** MPN value of water quality in Purwomartani Housings, Kalasan, Sleman DIY (data taken in April 2018).

|                | Average MPN value/ 100 ml |
|----------------|---------------------------|
| **MPN value/ 100 ml** |                         |
| Upstream 1     | 120                       | 57.34 ± 54.35 a          |
| Upstream 2     | 29                        |                           |
| Upstream 3     | 23                        |                           |
| Downstream 1   | 24                        | 808± 678.96 a            |
| Downstream 2   | 1200                      |                           |
| Downstream 3   | 1200                      |                           |

* variables following numbers with the same letter means the difference is not statistically significant.
Results showed that the mean MPN values of consecutive upstream and downstream points were 57.34 ± 54.35 /100 ml and 808 ± 678.96 / 100 ml. This value mean that total coliform in downstream (after bio filter treatment) was higher than in the upstream. We also analysed the average MPN value by statistical test and it showed that the treatment of water hyacinth as a bio filter in this study were not significantly different. Thus it can be said that the used of water hyacinth as bio filter did not affect the water microbiological quality.

To confirm the MPN test, we continued the positive results in presumptive MPN test to confirmed and completed test. The confirmed tests were done by inoculating the positive results in lactose broth from presumptive tests to the Endo Agar medium. The confirmed test showed positive results which appear to be greenish or metallic luster in Endo Agar Medium (Figure 2a). The colonies were then further tested using Gram stain for the complete test. Based from the staining test, it shown to be positive as coliform since it shown red colour with rod shape (Figure 2b). Thus, all the positive results in MPN tests were positive to be coliform.

![Figure 2. a. Coliform colonies shown in Endo Agar plate. Colony appear as greenish and metallic luster. b. Coliform cells indicated by red colour with rod shapes when stained using Gram stain](image)

The ineffectivity of water hyacinth as bio filter was probably affected by the surface area of the gravel and total root surface of the water hyacinth in the bio filter systems. It was probably expected that the surface area of bio filter was not quite large, also the gravel and the root surface were not so deep while the stream current was quite fast. Therefore solid suspension from the stream could not settled on the surface of the gravel and it reduced the chance for the microbial to attach on the surface of gravel and plant roots to make biofilm. This correlation also explained by [8] about factors influenced reduction of fecal coliform, that include greater system volume without increase in wetland surface area, retention time, increased surface area of the gravel, and greater total root surface area.

Another study from [15] reported that design of biofilter from plant also affect the effectivity of coliform removal in water. The selection of plant type may have an impact directly on removal process by root uptake, rhizosphere competition and predation, and release on antimicrobial exudates [10]. Water hyacinth were reported to have antimicrobial activites, including against E.coli. However, it was reported that the antimicrobial activity found in roots part was much lower than leaves part [16]. Therefore, the ineffectivity of water hyacynth as biofilter was probably related with this reason.

In addition, the existence of submerge zone in bio filter systems also plays important roles in retaining the water. This retained water in submerge zone may be processed during dry weather by increasing de-nitrification, maintain plant life and media consistency. With this process, it may alters the E.coli removal by adsorption and natural competition [15]. Therefore more research about design of biofilter from plants especially using water hyacinth should be conducted more advanced in future in order to optimize the ability of water hyacinth to reduce the number coliformin water sources.
4. Conclusion
Based on the discussion above, it can be concluded that the MPN values of coliform bacteria in the upstream and downstream points of Purwomartani Housing, Sleman District, DIY were 57.34 ± 54.35 /100 ml and 808 ± 678.96 / 100 ml. Statistical tests showed that the difference between before and after the addition of bio filter treatment was not significant so that the water hyacinth bio filter was not effective in microbiological control of water.

5. References
[1] Viala E 2008 *Irrig. Drainage Syst.* **22** 127–129
[2] Pandey P, Kass P, Soupir M, Biswas S, Singh, V 2014 *AMB Express*, **4** 51
[3] Hadiyanti A, Wibisono B 2012 *Tata Loka*. **14** (4) 295-303
[4] Jiuhui Q U 2008 *J. Env. Sci.* **20**(1) 1–13
[5] Ambashta R D and Sillanpää M 2010 *J Hazard Mater*. **180**(1-3) 38-49
[6] Geise G M, Lee H S, Miller D J, Freeman B D, Mcgrath J E, Paul D R 2010 *J Poly. Sci B: Polymer Physics* **48** 1685–1718
[7] Ghebremichael K A, Gunaratna K R, Henriksson H, Brumer H, Dalhammar G 2005 *Water Res.* **39**(11):2338–44
[8] Yongabi K.A 2010 *I.RE.CH.E* **2** (3) 444-458
[9] Coleman J., Hench K., Garbutt K., Sexstone A, Bissonnette G, Skousen J 2001 *Water. Air. Soil Poll.** 128 283
[10] Chandrasena G I, Shirdashtzadeh M, Li Y L, Deletic A, Hathaway J M, McCarthy D T 2017 *Ecol Eng* **102** 166-177
[11] Tulika T and Mala A 2015 *J. Plant. Sci.* **3**(1-1) 10-18
[12] Sindhu R, Binod P, Pandey A, Madhavan A, Alphonsa JA, Vivek N, Gnansounou E, Castro E, Faraco V 2017 *Bioresour Technol*. **230:**152-162
[13] APHA 2005 *Standard Method for Examination of Water and Wastewater*. 21 th ed. (Washington DC: America Public Health)
[14] Edberg S C, Rice EW, Karlin RJ, Allen MJ 2000 *Symp Soc Appl Microb* **5**(29) 106–116
[15] Chandrasena, G I, Pham T, Payne EG, Deletic A, McCarth 2014 *J.Hydrol*. **519** 814-822
[16] Fareed M F, Haroon A M, Rabeh S A 2008 *PJAS* **11** (21) 2454-2463

Acknowledgements
We thank to Indonesian Ministry of Research, Technology and Higher Education for Legal State University Assistance Fund-under the scheme of Research Grant on Collaboration of Lecturer and Students, Faculty of Biology Universitas Gadjah Mada Indonesia (grant number UGM/BI/1681/M/05/01) for supporting this project.