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Response of Lettuce (*Lactuca sativa* L.) To Aquaculture Wastewater Treatment

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Abstract

The fish feed waste and litters are organic matter that can be a source of nutrients for plants. This study is aimed to determine the growth and production of lettuce (*Lactuca sativa* L.) treated with wastewater from hard-lipped barb culture of several population densities. The research was carried out from April through June of 2020 at the greenhouse of the Agrotechnology Study Program, Faculty of Agriculture, Djuanda University, Bogor, Indonesia. The controlled study used a randomized block design with one factor, wastewater from fish population density of 10, 20 and 30 per 21 L of water, and AB Mix as a control. Plants treated with the AB Mix were significantly superior to those plants treated with the hard-lipped barb farming wastewater, demonstrated by taller plants, more and larger leaves, larger stem diameter and leaf area, longer roots, greater fresh and dry weight, as well as a higher leaf nitrate content. The aquaculture wastewater at the tested fish density in this study did not support optimal growth of lettuce, so lettuce production is still very low compared to those grown with the AB mix.

Keywords: hard-lipped barb, C/N ratio, nutrition

Introduction

Vegetables are consumed by 97.29% of the total Indonesian rural and urban communities (BPS, 2017). Vegetables are a source of vitamins, minerals, dietary fibers, and antioxidants that are all important for human health. Lettuce (*Lactuca sativa* L.) is one of the leafy vegetables belonging to the Asteraceae family with a very high economic value after cabbage, cauliflower, and broccoli (Wardhana et al., 2016).

According to the Central Statistics Agency (2017), vegetable crop production in Indonesia in 2014-2015 had positive developments, especially in the lettuce, with a total production of to 290,993 tons in 2014, an increase of 15.64% to 336,494 tons in 2015.

The demand for vegetables is increasing with the growing population, but agricultural land is shrinking due to the conversion of productive agricultural land into residential land and factory buildings, or housing. It is important to develop cultivation techniques, such as hydroponics, that are not completely dependent on ordinary agricultural land.

The advantages of hydroponic farming systems include more controlled plant environment, the uses of sterile planting media, efficient uses of water and fertilizers, protection from rain and direct sunlight so that the crops can grow in all seasons (Silvina and Syafrinal, 2008). Substrate hydroponics is a plant culture method that uses soilless media so that the roots grow on a porous substrate supplied with nutrient solution requirements (Nelson, 2009). AB Mix is a commercial source of the nutrients that are usually used in hydroponic culture. The chemical fertilizers are required for plant growth, but the excess can be detrimental to the environment. On the other hand, the fertilizer prices are increasingly expensive. The wastewater from fish feeds and manures can be a potential source of nutrients for crops, particularly nitrogen, in the form of ammonia, nitrites, and nitrates (Darmawan, 2010). The uses of aquaculture wastewater could potentially reduce the discharge of effluents into water environment.

Hard-lipped barb is a freshwater fish endemic to Indonesia which has high nutritional values. High density fish farming increases nitrogen in the wastewater, which comes from the accumulation of left-over feeds and fish feces (Darmawan, 2010). Our study was conducted to determine the growth and production of hydroponic lettuce treated with wastewater from hard-lipped barb culture at various fish population densities.

Materials and Methods

Location

The research was carried out from April through June of 2020 at the experimental greenhouse of the Agrotechnology Study Program, Faculty of Agriculture, Djuanda University, Bogor. The temperature in the greenhouse ranged from 28-33°C with the light intensity ranging from 6,833 to 24,967 lux.

Materials

The study used ”Green Curly” lettuce seeds, seedling trays, 45-L buckets, 50 x 85 cm and 60 x 100 cm PE plastics, measuring cups (1 L and 50 mL), calipers, analytical balances, hygrometers, lux meters, pH-meter, TDS-meter, and 32 aquarium measuring 30 cm x 30 cm x 30 cm. The materials used include rockwool, 30 cm x 30 cm polybags, husk charcoal, and wastewater from hard-lipped barb (*Osteochilus hasseltii* CV) culture.

Experimental Design

The study was arranged in a randomized block design with a single factor, i.e. the density of bonylip barb culture in its wastewater, i.e. 10, 20, and 30 fishes per 21 L of water, and a control of AB Mix (5 ml/liter of water). Each aquarium was treated with 250 mL of wastewater, or AB Mix solution.

The research was started by sowing lettuce seeds in a 3x3 cm rockwool media with a 5 mm sowing depth. Aquaculture wastewater was obtained by siphoning an aquarium containing lipped barbs. The AB Mix was in the form of crystals which were prepared as a stock solution.

Lettuce seedlings were transplanted at 15 days after sowing. The planting medium used was husk charcoal, which was put into 30x30 cm polybags, or about 1.4 kg per polybag. Plants were watered every morning and evening every day. Replanting was conducted on the plants that did not grow and was conducted at 7, 14, and 21 days after planting (DAP). Weeding was conducted fortnightly by manually pulling weeds that grew in and around the polybags of plants. Pest and disease control was carried out manually without the use of pesticides to avoid contamination of plants from harmful chemicals. Lettuce was harvested at about 42 DAP.

The growth measurements were conducted weekly recording the number of leaves that were fully opened, the plant height, the leaf length, leaf width, the stem diameter and also the leaf area from 7 DAP to 42 DAP. Plant height was measured from the base of the stem to the growing point using a ruler. The leaf length was measured from the base of the leaf to the tip of the leaf, whereas leaf width was measured from side to side of the widest leaf following the radius of the leaf segment using a ruler or tape measurement. Lettuce stem diameter was measured each week from 21 to 42 DAP using a caliper. Leaf area was measured on the 5th widest leaf from the shoot using the gravimetric method. Leaf nitrate was measured using a nitrate meter (Horiba). Root lengths, shoot, and root fresh and dry weight were measured using a digital scale after drying. The chlorophyll content analysis was carried out at the Laboratory of the Department of Agronomy and Horticulture, Faculty of Agriculture, Bogor Agricultural University.

Data was analyzed by analysis of variance at 5% level using SPSS; further test was conducted using Duncan’s Multiple Range Test (DMRT) at 5% level.

Results and Discussion

Chemical and Physical Properties of The Wastewater

The fish culture wastewater had a pH higher than the AB mix, and a EC lower than the AB mix at the same temperatures (Table 1). Amongst the wastewater, the ammonia content from the wastewater with a fish population density of 30 was the highest (2.21 ppm), followed by a density 20 at 1.99 ppm and density of 10 (0.95 ppm, Table 1). A high ammonia content in the water can inhibit fish growth. A study by Pitrianingsih et al. (2014) demonstrated that water ammonia concentration >0.3 mg/L resulted in fish with decreased appetite, and even caused the fish to die. However, for plant culture, the higher the ammonia, the more nitrogen is available for the plants. For example, Damayanti et al. (2018) reported that the addition of ammonium (NH_4^+) and nitrate (NO_3^-) increased plant height, the number of leaves, the leaf area, and the fresh weight. Therefore, the system developed here has the potentials to create a symbiotic mutualism between the fish and the crops.

Rockwool can hold 92% of water so it can store nutrients for a long time (Agustina, 2019). Husk charcoal has good porosity which was a beneficial property for aeration and drainage so that plant roots could absorb nutrients optimally (Perez, 2008).

The lettuce in this study survived under high temperatures of the nutrient solution and the greenhouse. According to Priandoko et al. (2000) the optimal temperature of nutrient solution is 12-22°C, however, in this study it was 27.3°C, whereas the

Table 1. Chemical and physical properties of the fish culture wastewater at several population density

Parameter	AB Mix	Density 10	Density 20	Density 30
Ammonia (ppm)		0.95	1.99	2.21
pH	7.4	8.4	8.6	8.5
EC (ppm)	1888	439	469	485
Temperature (°C)	27.2	27.2	27.2	27.3

greenhouse temperature was 28-33°C. The optimum temperature for lettuce growth was 15-25°C (Amalia, 2013; Aini et al., 2010). According to Fariudin et al. (2017) leaf lettuce was relatively able to adapt to high temperatures in the lowland.

Lettuce Height and Leaf Number

At 7-42 DAP the lettuce height treated with AB Mix was significantly taller those treated with lipped barb farming wastewater (Table 2). There was no significant differences in the plant height treated with wastewater of different fish densities (Table 2).

Leaf Size

Lettuce leaf size with the AB Mix treatment was larger than the those treated with lipped barb wastewater (Table 4). A study by Perwitasari et al. (2012) reported that higher nitrogen promoted leaf expansion. Leaf length of the plants in different wastewater treatments were not significantly different from 13 to 42 DAP (Table 4).

Table 4. Lettuce leaf length and leaf width at 14, 28 and 42 days after planting (DAP) after treatment with AB Mix and fish culture wastewater at several population density.

Table 2. Lettuce plant height at 7 to 42 days after planting (DAP) after treatment with AB Mix and fish culture wastewater at several population density

Treatment	Plant height (cm)					
	7 DAP	14 DAP	21 DAP	28 DAP	35 DAP	42 DAP
AB Mix (control)	9.48[c]	11.99[b]	16.73[b]	22.93[b]	35.16[b]	43.74[b]
Density 10	7.16[a]	9.03[a]	12.21[a]	15.90[a]	21.40[a]	23.63[a]
Density 20	8.17[b]	9.80[a]	12.64[a]	17.19[a]	22.84[a]	25.38[a]
Density 30	7.86[ab]	9.49[a]	12.53[a]	16.32[a]	21.77[a]	24.39[a]

Note: The average values in the same column followed by the same letter are not significantly different according to the DMRT at the 5% level.

The wastewater treatment did not significantly affect the number of leaves at 7 to 14 DAP. It is possible that at the beginning of growth the amount of nutrients available were sufficient for plant growth, but became deficient at later stages (21-42 DAP). The AB mix contain macro nutrients and micronutrients, while the nutrients contained in the wastewater of the lipped barb cultivation consisted of only nitrogen. A study by Muhadiansyah et al. (2016) reported that lettuce that was grown without micronutrients Zn, Mo, Fe, Mn, Co and B had fewer leaves than those supplied with micronutrients.

At 21 DAP, however, the number of leaves on the plants treated with the AB Mix were not significantly different from those treated with wastewater with a fish density of 30 (Table 2). The nitrogen content in the AB Mix with the wastewater at a density of 30 was higher than in wastewater at a density 20 and 10, indicated by a high ammonia content (Table 1).

In a study by Meriyanto et al. (2017) N concentration of 900 ppm is optimum for crop growth, and that N is the most important nutrient for promoting plant stem and leaf growth.

The pH value determines the availability of the nutrients. At an alkaline pH, the N availability decreases hence disrupt the roots function to absorb nutrients (Fariudin et al., 2017). In addition, the alkaline environment reduced the activity of microorganisms that decompose organic matter (Saputra et al., 2016). According to Megasari (2017) a neutral pH is optimal for crop growth.

AB mix has a total nutrient of 1888 ppm with a pH of 7.4, whereas the wastewater from fish population density of 10, 20 and 30 had 439 ppm, 469 ppm, and 485 ppm, respectively, with a pH of > 8 (Table 1). This points to the lower quality of the lettuce production,

Table 3. Lettuce number of leaf at 7 to 42 days after planting (DAP) after treatment with AB Mix and fish culture wastewater at several population density

Treatment	Number of leaves					
	7 DAP	14 DAP	21 DAP	28 DAP	35 DAP	42 DAP
AB Mix (control)	4.44	5.25	6.56[c]	11.50[b]	22.94[b]	31.63[b]
Density 10	4.50	5.00	5.75[a]	8.19[a]	13.38[a]	16.63[a]
Density 20	4.25	4.94	6.00[ab]	8.06[a]	13.31[a]	16.13[a]
Density 30	4.19	5.13	6.31[bc]	8.81[a]	14.63[a]	17.75[a]

Note: The average values in the same column followed by the same letter are not significantly different according to the DMRT at the 5% level.

Table 4. Lettuce leaf length and leaf width at 14, 28 and 42 days after planting (DAP) after treatment with AB Mix and fish culture wastewater at several population density.

Treatment	Leaf length (cm) at			Leaf width (cm) at			Stem diameter (mm)	Leaf area (cm^2)
	14	28	42	14	28	42	42	42
	(days after planting)							
AB Mix (control)	11.16[b]	18.41[b]	22.84[b]	7.93[b]	18.89[b]	27.01[b]	16.56[b]	122.96[b]
Density 10	7.84[a]	12.18[a]	14.27[a]	4.43[a]	7.04[a]	8.16[a]	5.06[a]	38.17[a]
Density 20	8.44[a]	12.10[a]	15.03[a]	4.50[a]	6.13[a]	8.47[a]	4.78[a]	37.75[a]
Density 30	8.36[a]	12.16[a]	13.71[a]	4.64[a]	6.61[a]	7.96[a]	4.56[a]	39.22[a]

Note: The average values in the same column followed by the same letter are not significantly different according to the DMRT at the 5% level.

indicated by smaller leaf and stem diameter compared to those treated with the AB mix (Table 4).

Plant Productivity

Root length, shoot and root fresh and dry weight were significantly reduced when treated with wastewater (Table 5).

The vegetative growth rate was synergistically related to root length, fresh weight, and dry weight. Long dan fibrous roots indicate the amount of nutrients and the amount of water absorbed. In the wastewater treated plants it is suspected that a lot of the nutrients and water were lost due to the evaporation as the temperature in the greenhouse was quite high. The evaporation from the AB Mix treatment was presumably less as the large leaves shaded each other. Greater fresh shoot weights are related to the optimal root growth that supported the function of absorbing nutrients and water (Fariudin et al., 2017).

Lettuce Leaf Quality

The quality of lettuce in this study was based on the leaf content of nitrate and chlorophyll. Lettuce nirate was the highest with AB Mix, and there was no significant differences in the nitrate and chlorophyll levels with different wastewater treatments (Table 6).

The role of chlorophyll is to absorb light for photosynthesis. One of the factors that affects the chlorophyll content of a plant is leaf morphology (Setiari and Yulita, 2009). The chlorophyll content of the lettuce in this study was not affected by the AB Mix or the wastewater treatment, likely due to the thin nature of the lettuce leaves. According to Rahmi (2017) chlorophyll compared to kale, green spinach, and mustard green leaves. In lettuce with AB Mix treatment, large leaf areas did not have a positive correlation to the chlorophyll content.

Nitrogen is absorbed by plants in the form of ammonium (NH_4^+) and nitrate (NO_3^-) through several stages of the nitrogen cycle. Nitrogen cycle is a process of converting nitrogen into various other chemical forms; starting from N fixation, ammonification, nitrification, and denitrification. The successfully absorbed nitrate was reduced back to nitrogen gas (N_2), known as denitrification, and then released into the air to complete the nitrogen cycle, and this cycle continues as long as the plant grows (Damayanti et al., 2018).

Ammonia is converted to ammonium in the

Table 5. Fresh weight and dry weight of lettuce shoots and roots treated with fish culture wastewater at several population density.

Treatment	Shoot			Root		Total Shoot + Root	
	Root length (cm)	FW (g)	DW (g)	FW (g)	DW (g)	FW (g)	DW (g)
AB Mix (control)	38.95[b]	447.53[b]	21.13[b]	32.66[b]	3.23[b]	240.09[b]	12.18[b]
Density 10	10.88[a]	14.36[a]	1.36[a]	0.59[a]	0.12[a]	7.48[a]	0.74[a]
Density 20	14.10[a]	21.60[a]	1.40[a]	0.55[a]	0.12[a]	11.08[a]	0.76[a]
Density 30	11.73[a]	27.00[a]	1.72[a]	1.03[a]	0.18[a]	14.01[a]	0.96[a]

Note: The average value in the same column followed by the same letter is not significantly different according to the DMRT at the 5% level.

Table 6. Lettuce leaf nitrate and chlorophyll contents treated with fish culture wastewater at several population density.

Treatment	Nitrate (ppm)	Chlorophyll a (mg/g)	Chlorophyll b (mg/g)
AB Mix (control)	7791.67[b]	0.90	0.43
Density 10	4175.00[a]	0.89	0.44
Density 20	4383.33[a]	0.89	0.44
Density 30	3950.00[a]	0.93	0.45

Note: The average value in the same column followed by the same letter is not significantly different according to the DMRT at the 5% level.

ammonification process. If conditions are favorable, the nitrification process will occur. Nitrification is the conversion of ammonium to nitrate by involving microorganisms. Factors that affect the nitrification process include planting media, oxygen availability, temperature, and pH. Lettuce in this study was grown on husk charcoals which is a highly porous substrate, thus provided good aeration that supports nitrification.

The average temperature of the greenhouse during the research was relatively high, i.e., 28-33°C and the temperature of the nutrients can reach 27.3°C. Our study demonstrated that lettuce can still grow at these temperature ranges. High temperatures and alkaline pH can inhibit the activity of microorganisms in the nitrification process. Bacteria can still grow and reproduce at a pH of 6.5-7.5 (neutral) with temperatures of 25-35°C (Anisa and Welly, 2017). The nitrate in the fish culture wastewater was the end product of the ammonia oxidation, assisted by *Nitrobacter* sp, which was then then utilized by the plants for growth. The low nitrate levels in the wastewater indicates that the oxidation of the ammonia was slow, presumably due to the inhibition of the microbial activities and too alkaline water.

Conclusion

Plants treated with the AB mix grew significantly better and had a higher leaf nitrogen content than those treated with the aquaculture wastewater. The aquaculture wastewater at the tested fish density in this study did not support optimal growth of lettuce, so lettuce production is still very low compared to those grown with the AB mix.

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References

Agustina, R. (2019). "The Effect of Hydroponic Media and Nutrient Composition on the Growth of Green Lettuce Leaves (*Lactuca sativa* L.)" [Thesis]. Universitas Malikussaleh, Aceh.

Aini, R.Q., Yaya, S., Muhammad, N.H. (2010). Application of KPD bionutrients to curly lettuce (*Lactuca sativa* L.). *Jurnal Sains dan Teknologi Kimia* **1**, 73-79.

Amalia, D.,R.,N. (2013). "Efek Temperatur terhadap Pertumbuhan *Gracilaria verrucosa".* [Thesis]. Universitas Jember. Jember

Anisa, A., and Welly, H. (2017). Pengolahan limbah domestik menggunakan moving bed biofilm reactor (MBBR) dengan proses aerobik-anoksik utuk menurunkan nitrogen. *Jurnal Teknik ITS* **6**, F361-F366.

Badan Pusat Statistika. (2017). "Konsumsi Buah dan Sayur Susenas Maret 2016". Kementerian Pertanian, Jakarta.

Damayanti, D.,P.,O., Tri, H.,and Slameto. (2018). Pengaruh ammonium (NH_4^+) dan nitrat (NO_3^-) terhadap pertumbuhan dan kandungan minyak atsiri tanaman kemangi (*Ocimum basilicum*) dengan sistem hidroponik. *Agritrop* **16**,163-175.

Darmawan, W.,P.,J. (2010). "Pemanfatan Air Buangan Lele Dumbo sebagai Media Budidaya *Daphnia* sp." [Thesis]. Bandar Lampung: Universitas Lampung.

Fariudin, R., Endang, S., and Sriyanto, W. (2012). "Pertumbuhan dan Hasil Dua Kultivar Selada (*Lactuca sativa* L.) dalam Akuaponik pada Kolam Gurami dan Kolam Nila" [Thesis]. Universitas Gadjah Mada. Yogyakarta

Fitriasari, N. (2020). "Pengaruh Komposisi Pupuk Urea dan Kipahit terhadap Pertumbuhan, Produksi dan Kualitas berbagai Aksesi Kemangi (*Ocimum basilicum* L.)" [Thesis]. Universitas Djuanda, Bogor.

Megasari, R. (2017). "Teknologi Aquaponik Tanaman dan Ikan Nila pada Tiga Jenis Media Tanam dan Frekuensi Pemupukan" [Thesis]. Universitas Hasanuddin. Makassar.

Meriyanto, Busroni, A., and Sari, A. (2017). Pengaruh pemberian berbagai konsentrasi larutan nutrisi hidroponik terhadap pertumbuhan dan hasil tanaman selada merah (*Lactuca sativa* L.) dengan sistem deep flow technique (DFT). *Jurnal Triagro* **2**, 28-37.

Muhadiansyah, T.,O., Setyono, and Sjarif, A.,A. (2016). Efektivitas pencampuran pupuk organik cair dalam nutrisi hidroponik pada pertumbuhan dan produksi tanaman selada (*Lactuca sativa* L.). *Jurnal Agronida* **2**, 37-46.

Nelson R. (2009). Methods of hydroponic production. *Journal of Aquaponics* **4**, 24-30.

Perez LE. 2008. "Hydroponics for The Home". Inter-American Institute for Cooperation on Agriculture. San Jose.

Perwitasari, B., Mustika, T., Catur, W. (2012). Pengaruh media tanam dan nutrisi terhadap pertumbuhan dan hasil tanaman pakchoi (*Brassica juncea* L.) dengan sistem hidroponik. *Jurnal Agrovigo* **5**, 4–25.

Pitrianingsih,C., Suminto, and Sarjito. (2014). Pengaruh bakteri kandidat probiotik terhadap perubahan kandungan nutrien C, N, P dan K media kultur lele dumbo (*Clarias gariepinus*). *Journal of Aquaculture Management And Technology* **3**, 247-256.

Priandoko, A.,D., Parwanayoni, S.,and Sundra, I.,K. (2000). Kandungan logam berat (Pb dan Cd) pada sawi hijau (*Brassica rapa* L. Subsp. Perviridis Bailey) dan wortel (*Paucus Carrota* L. var. *sativa hoffim*) yang beredar di kota denpasar. *Jurnal Simbiosis* **1**, 9-20.

Purnomo, J., Dwi, H., and Trijono, D.,S. (2016). Budidaya cabai rawit sistem hidroponik substrat dengan variasi media dan nutrisi. *Journal of Sustainable Agriculture* **31**,129-136.

Rahmi N. (2017). "Kandungan Klorofil pada Beberapa Jenis Tanaman Sayuran sebagai Pengembangan Praktikum Fisiologi Tumbuhan" [Thesis]. Universitas Islam Negeri Ar-Raniry. Bamda Aceh.

Saputra, A.D., Haeruddin, and Niniek, W. (2016). Efektifitas kombinasi mikroorganisme dan tumbuhan air lemna minor sebagai bioremediator dalam mereduksi senyawa amoniak, nitrit dan nitrat pada limbah pencucian ikan. *Diponegoro Journal of Maquares* **5**, 80-90.

Setiar,i N., Yulita, N. (2009). Eksplorasi kandungan klorofil pada beberapa sayuran hijau sebagai alternatif bahan dasar food supplement. *Bioma* **11**, 6-10.

Silvina, F., Syafrinal. (2008). Penggunaan berbagai medium tanam dan konsentrasi pupuk organik cair pada pertumbuhan dan produksi mentimun Jepang (*Cucumis sativus*) secara hidroponik. *Jurnal Sage* **7**, 7-12.

Siregar, J., Sugeng, T., and Diding, S. (2015). Pengujian beberapa nutrisi hidroponik pada selada (*Lactuca Sativa* L.) dengan teknologi hidroponik sistem terapung (THST) termodifikasi. *Jurnal Teknik Pertanian Lampung* **4**, 65-72.

Wardhana, I., Hudaini, H., Insan, W. (2016). Respon pertumbuhan dan produksi tanaman selada (*Lactuca sativa* L.) pada pemberian dosis pupuk kandang kambing dan interval waktu aplikasi pupuk cair super biotik. *Agritrop Jurnal Ilmu Pertanian* **14**, 165-185.

Yama, D.I., Hendro, K. (2020). Pertumbuhan dan kandungan klorofil pakcoy (*Brassica rappa* L.) pada beberapa konsentrasi AB *Mix* dengan sistem wick. *Jurnal Teknologi* **12**, 21-30.