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To cite this article: A Satya et al 2020 IOP Conf. Ser.: Earth Environ. Sci. 535 012027

View the article online for updates and enhancements.

doi:10.1088/1755-1315/535/1/012027

Nitrogen uptake competition between minute duckweed (*LemnaperpusillaTorr*) and microalgae under various nutrient composition

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Abstract. Implementation of minute duckweed (Lemnaperpusilla) in an integrated aquaculture system for encouraging movement of in-lake floating cage aquaculture to inland base aquaculture has been emphasized as one among various measures for restoring the degraded Lake Maninjau water quality. Unfortunately, in the field, there is a harmful competition between microalgae and duckweed for nutrient uptake. This paper reports an attempt to find out suitable nutrient solution composition for supporting duckweed growth while suppressing the microalgal proliferations. Batch culture of the plant with six varied media treatments (A to L) whichmade from Nitrogen- Phosphorus-Potassium (NPK) fertilizers basis enriched with trace elements, bio-algaecide (Terminalia Cattapa extract), and molasses solutions were examined. The adsorption of TN by duckweed and the assemblage of natural microalgae community was quantified and modelled using mass balance concept approach and non-linear equations. The result showed that among of 12 obtained models, treatment E (a solution which made from NPK fertilizer enriched with molasses and trace element) was the most favourable media composition for promoting duckweed growth and simultaneously suppressed microalgae growth. It is proven from the fact that the TN uptake patterns and the chlorophyll-a content which show chlorophyll-a content werethree magnitudes higher than those were found in treatment A (the most optimum medium for growing microalgae community). The TN uptake rate by minute duckweedin treatment of E was observed as much as 251.54 mg/m²/day while that ofmicroalgae was 208.60 mg/m²/day.

1. Introduction

Nitrogen is one of the fundamental elements for aquatic biota. Excessive loading of this element into the lake, however, can drive to destructive ecological condition, such aswater eutrophication [1, 2]. The actual example of that situation occurs in Lake Maninjau waters in the West Sumatra province. This lake water body has been suffered hyper-eutrophication symptom due to the excessive internal nutrient loading came from massive uncontrolled floating fish cage activities [3, 4]. Accordingly, it is necessary to reduce the nutrient input by means of remove aquaculture activities from the lake water body and finding out the alternative of amore sustainable sound aquaculture system [5, 6].

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doi:10.1088/1755-1315/535/1/012027

The minute duckweed (*Lemna perpusilla*) is widely known to have high productivity (13–38 tons dry weight/hectare/year) and nutritioncontent (as protein) up to 6–10 magnitudes than of soybean [7]. This plant can be grown in relatively high of nitrogen, phosphorus and potassium (NPK) concentration medium and accumulate those minerals for manufacturing biomass [8]. It has low fibre content while contains high protein and rich invarious essential amino acids, so that can be digestedeasily when it is used foranimal feed. Protein and carbohydrate in *Lemna minor* grown in organic manure was 36.07±0.18% and 34.07±0.36%, but when grown in inorganic fertilizer, the protein and carbohydrate are 27.12±0.40% and 46.31±0.74% [9]. Therefore, this plantis prospective in supporting the sustainable aquaculture, such as through an integrated aquaculture system proposed by RC for Limnology which combines minute duckweed with Nile tilapia (*Oreochromis niloticus*) cultures at the area nearbyLake Maninjau. This effort is a part for tackling the Lake Maninjau problems which is directed toimprovethe lake water quality and searching for more environmentally sounds activity for earning life. Unfortunately, implementation of duckweed culture in the field frequentlyhindered by nutrient uptake competition with the existingnatural plants, in which mostly minute duckweed isoutcompeted by the existing microalga community [3, 10].

Nutrient uptake competition between duckweed culture and microalgae in an experimental pond is rarely reported, even it commonly occurs in the natural situation [11, 12]. It is widely known that different processes related to the aquatics biological-chemical-and physical characters play an important role in nutrient uptake, both in natural and artificial conditions. However, the artificial situation is more controllable than natural condition so that general pattern can be more easily obtained [13, 14]. Accordingly, a study focused on the nutrient uptake pattern involving minute duckweed and microalgae in laboratory scale is necessary to be conducted. This study attempted to find out a suitable nutrient composition for supporting duckweed growth while suppressing the microalgae proliferation. That projected situation was determined from the shape of nitrogen uptake pattern during the time course of the experiment, as well comparison uptake rate between minute duckweed and microalgae. Furthermore, those data should be supported by obtained higher growth rate and biomass productivity of the duckweed.

2. Materials and Methods

2.1. Experimental design

Lemna perpusilla in this experiment was obtainedfrom culture pond in RC for Limnology-LIPI Cibinong, while microalgae community was grown naturally. This paper only focuses on the discussion of total nitrogen dynamic; therefore, the structure of microalgae community (e.g knowing on microalgae species) is not discussed. The experiment was conducted on a batch system in plastic boxes of 0.27 x 0.35 m² with working volume of 10 L, placed in a green house. Inorganic fertilizer of NPK (Nitrogen, Phosphorus, and Potassium) diluted with tap water was used as media solution basis, which was varied and enriched with bio-algaecide (*Terminalia cattapa* extract, added 10 mL/L), trace element (0.1 mL/L), and molasses (1 mL/L) solutions. Schematically, the designed process is described in figure 1.

doi:10.1088/1755-1315/535/1/012027

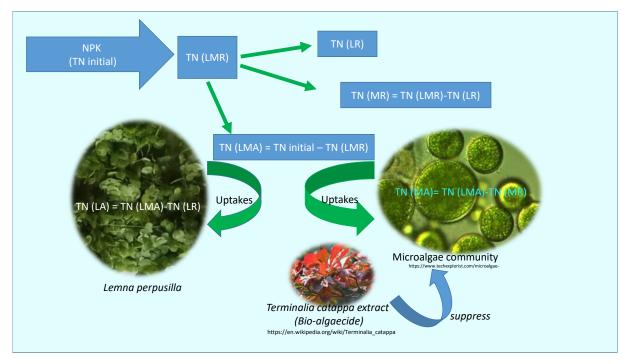


Figure 1. Hypothetical process of Total Nitrogen uptake in this designed observed system.

Remarks: TN (LMR)= TN remaining after adsorbed by Lemna & microalgae, TN (LR)= TN remaining after adsorbed only by Lemna, TN (MR)= TN remaining after only adsorbed by microalgae, TN (LMA)= TN adsorbed by Lemna & microalgae, TN (LA)= TN adsorbed only by Lemna, TN (MA)= TN adsorbed only by microalgae

The treatment of lpha was designed to vary the NPK of 100 mg/L with the addition of cattapa extract, trace element, and molasses solutions while treatment of β not only combinedwith trace element solution but also varied the NPK into doublet and triplet (as designed in table 1). Every treatment box was duplicated for a total 12 experimental units. Initially, a 10 g of minute duckweed biomass was stocked and let to grow from 13 February to 27 March 2019. Coincident with that, the existing wild microalgae community then naturally remained grew along with the experiment. There was no microalgae biomass was added. The initial biomass was adjusted to cover the total surface of the experimental box. Evaporative loss was compensated by adding tap water to the marked line made in container before the experiment commenced. The biomass and water were sampled every four days. After each of sampling, samples were stored inside of refrigerator at 4°C until analysis within one week of sampling date.

Table 1. Setting up of nutrient experiment using NPK in every box correspond to the designed treatment of α and β .

Box	Treatment of ά	Box	Treatment of β
A	NPK 100 mg/L (control)	G	NPK 100 mg/L (control)
В	NPK 100 mg/L+Cattapa extract	Н	NPK 100 mg/L+Trace Elements
C	NPK 100 mg/L+Trace Elements	I	NPK 200 mg/L
D	NPK 100 mg/L+Trace Elements+Cattapa extract	J	NPK 200 mg/L+Trace Elements
E	NPK 100 mg/L+Trace Elements+Molasses	K	NPK 300 mg/L
F	NPK 100 mg/L+Trace Element+Molasses+Cattapa extract	L	NPK 300 mg/L+Trace Elements

doi:10.1088/1755-1315/535/1/012027

2.2. Physicochemical characteristic of water

Water conductivity, pH, and turbiditywere directly measured during sampling by portable equipment (Lutron YK-2001PH, Lutron pH 201, Lutron TU 2016, Taiwan). Water samples were taken by unfiltered (for analysis of TN LMR), filtered through Whatman® Grade GF/C Glass Microfiber filters (for TN LR analysis) and the retained biomass was analysed for chlorophyll-a content which has a particle filtration size of 1.2 μ m according to Knefelkamp et al. [15]. Total nitrogen (TN) and chlorophyll-a were determined by persulphate digestion method followed by Brucine method and acetone extraction followed by three chromatic methods [16]for measuring with spectrophotometer Thermo ScientificTM GENESYS 10S, USA.

2.3. Data analysis

The relative grow rate (RGR) of minute duckweed (expressed as % / day) was calculated using the formula (1)

$$RGR = \ln\left(\frac{W_t}{W_0}\right)/tx100 \tag{1}$$

While the biomass productivity $(g/m^2/day)$ was calculated according to formula (2)

$$P = \frac{W_t - W_0}{t \times A}(2)$$

Where W_t and W_0 are the fresh weight of macrophyte at the time of harvest (t) and the time of introduction of plant, t is the time interval (days), A is the surface area of the box (m²).

While mass balance concept is approximated according to equation (3) to (6) in table 2.

Table 2. Mass balance approximation to calculate the remained and absorbed Total Nitrogen

Abbreviations	Remarks	Formula
TN (LMR)	TN remaining after adsorbed by Lemna &	
	microalgae	
TN (LR)	TN remaining after adsorbed only by Lemna	
TN (MR)	TN remaining after only adsorbed by microalgae	= TN (LMR)-TN (LR) (3)
TN (LMA)	TN adsorbed by Lemna & microalgae	= TN (initial)- TN (LMR) (4)
TN (LA)	TN adsorbed only by Lemna	= TN (LMA) - TN (LR) (5)
TN (MA)	TN adsorbed only by microalgae	= TN (LMA)-TN (MR) (6)

Uptake rate (q $_{TN}$) expressed as $mg/g.m^2.day$ (wet weight) (equation 7) and q $_{TN}$ expressed in $mg/g/m^2$ (equation 8).

$$q_{TN,wet\ weight} = \frac{(TN_{adsorbed} \times V_{working})}{(m \times A \times t)}$$

$$q_{TN} = \frac{(TN_{adsorbed} \times V_{working})}{(A \times t)}$$
(8)

 W_t represented the fresh weight of the duckweed(grams) collected at time of t (day) and initial duckweedbiomass (grams) which was added in the pond; A= surface area of the box(0.0945 m²); $TN_{adsorbed}$ = TN adsorbed by biomass (mg/L); $V_{working}$ = working volume=10 L.

Mean value was calculated for all analyzed parameters. The statistical test and curve fitting with non-linear equation models were performed using Sigma Plot 10.0. Normality test was conducted with Durbin-Watson. The distributed normally data were then tested with ANOVA followed by post hoc testing using Tukey's test. When significant differences between treatment were present, Mann-Whitney test were performed. During statistical tests, the level of significance was held at 0.05.

doi:10.1088/1755-1315/535/1/012027

3. Results and Discussion

3.1. Physicochemical characteristic of treatment pond

Table 3shows a comparison of the average initial physicochemical characters of 12 treatment boxes which containing nutrient solutions which were made as designed according to table 1. The total nitrogen (TN) in boxes which filled with NPK of 100 mg/L (treatment box of A to H) were ranged of 15.33 - 15.82 mg/L. Meanwhile, as NPK was doubled and tripled, TN concentrations were correspondingly of ranged 26.90 to 30.46 mg/L (treatment box of I and J) and 33.81 to 39.90 mg/L (treatment box of K and L). These observed TN values are reasonable since NPK used here was NPK Triple 16, which means that it contains 16% of N (Nitrogen), 16% of P₂O₅ (Phosphate), 16% of K₂O (potassium), 0.5% of MgO (Magnesium), and 6% of CaO (Calcium) [17]. Range of pH values were 5.14 to 7.47, which tended to be more acidic when NPK added in triple. Chlorophyll-a (chl-a) contents ranged of 139.83 mg/m³ to 767.64 mg/m³. The use of NPK 200 mg/L as a nutrient basis was promoted 2 to 5 magnitudes of chlorophyll-a content, but when NPK 300 mg/L was used, the chl-a was only higher of three folds. The conductivity of the solution (ranged of 0.163 to 0.450 mS/cm) also proportionate to the concentration of NPK. The adding of trace element solution and cattapa extract as bio-algaecide was lowered the conductivity. This experiment was conducted at a water temperature from 26.9 to 30.0°C, means that it was typical for tropical climate. Water turbidity ranged from 5.3 to 22.7 NTU. Treatment box of A to F were prone to be higher than of G to L. It was probably because the occurrence of the mixture of the NPK and organic substances (cattapa extract, molasses, and trace element) enable to support the microbial growth in the solution which lead to the increasing of turbidity, as also reported by other researchers [18]. On the contrary, more added three folds of NPK in treatment boxes of G to L reduced turbidity values of more than three folds.

Table 3. The average physicochemical data at the initial observation in every treatment box. Details of each media composition reffers to table 1.

Parameter	Treatment of ά						
1 arameter	A	В	C	D	E	F	
TN (mg/L)	15.78	15.33	15.74	15.29	15.70	15.53	
Chlorophyll-a (mg/m ³)	212.10	139.83	177.73	170.58	294.27	312.75	
pH	5.53	5.34	5.36	5.71	6.84	6.91	
Conductivity (mS/cm)	0.195	0.186	0.184	0.163	0.164	0.173	
Temperature (°C)	27.4	27.0	27.0	26.9	26.9	27.0	
Turbidity (NTU)	15.6	16.9	14.9	16.9	22.7	21.9	
Parameter	Treatment of β						
	G	Н	I	J	K	L	
TN (mg/L)	15.82	15.78	29.07	30.46	39.90	33.81	
Chlorophyll-a (mg/m³)	265.38	211.92	767.64	607.75	286.73	357.65	
pH	7.20	7.47	5.86	5.37	5.31	5.14	
Conductivity (mS/cm)	0.170	0.185	0.377	0.338	0.471	0.450	
Temperature (°C)	30.0	29.3	29.1	29.0	29.1	29.4	
Turbidity (NTU)	12.7	13.2	12.7	8.6	8.2	5.3	

doi:10.1088/1755-1315/535/1/012027

3.2. Growth and productivity of Lemna perpusilla

Minute duckweedhad highest specific growth rate (SGR) in the treatment box of L at 20.50 %/day, followed by treatment box of K at 19.94 %/day; thus Lemna was considered better adapted in medium with NPK concentration of 300 mg/L (table 4). These values were comparable with SGR of Lemna (18–23 %/day) grew on Lake Maninjau water as reported by Chrismadha et al [3]. However, boxes of A to F (the α treatment) were only slightly lower than boxes of K and L (the β treatment). It means that the adding NPK of 100 mg/L with various other added substances (bio-algaecide, trace element, and molasses) gave moderate satisfying SGR values. At the same time, lowest SGR was observed on box treatment of G followed by the box of H. This low SGR value might be attributed tosudden drop of pH values which occurred at the initial stage of cultivation and leaded to lowerbiomass produced afterwards.

The biomass productivity of theminute duckweedalso shows a similar pattern with those of SGR (table 4). The range of biomass productivity in wet weight was 16.40 to 40.38 g/m²/day, which slightly lower than that cultivated in an experimental ponds system (51 g/m³/day) nearby the Lake Maninjau [3], which might be in consequence of different culture conditions.

Table 4. The average growth and productivity of Lemnaperpusilla

Parameter *)	Treatment box of ά treatment						
Tarameter ')	A	В	С	D	Е	F	
SGR of <i>Lemn</i> a (%/day) Biomass productivity of <i>Lemna</i>	18.19	19.01	19.64	19.66	18.19	18.68	
(g/m²/day)	32.35	34.84	37.06	37.33	34.37	35.66	
Parameter *)	Treatment box of β treatment						
	G	Н	I	J	K	L	
SGR of Lemna (%/day)	7.58	8.50	17.08	16.46	19.94	20.50	
Biomass productivity of <i>Lemna</i>							
Biomass productivity of <i>Lemna</i> (g/m²/day)	16.40	18.16	33.42	33.16	40.21	40.38	

^{*)} in average wet weight

Table 5 presents the average uptake rate of TN by minute duckweedand microalgae community which estimated for55 days observation. The uptake rate of TN was expressed in two units, which are mg TN/g/m²/day and mg TN/m²/day, in order to evaluate the role of biomass production and covering surface area in determining the plant phytoremedial capacity, which further allows the estimation of phytoremediation potential based on the SGR value. In general, the treatment of α showed lower range of TN uptake rate (3.57 to 4.84 mg/g/m²/day or 191.89 to 251.54 mg/m²/day) than thetreatment β (2.14 to 5.49 mg/g/m²/day or 117.31 to 264.50 mg/m²/day). Similarly, the average of TN uptake rate by microalgae on the treatment of α had lower range (208.22 to 248.06 mg/m²/day) than the β treatment (117.66 to 663.07 mg/m²/day). The highest TN uptake rate by microalgae in the α treatment was on box C (NPK 100 mg/L added with the trace element solution) which was 267.61 mg/m²/day. In β treatment group,TN uptake rate by microalgae was almost three folds higher which the highest rate was found in box K (NPK 300 mg/L) followed by box L (NPK 300 mg/L enriched with trace element solution) which were 663.07 mg/m²/day and 608.91 mg/m²/day.

doi:10.1088/1755-1315/535/1/012027

Table 5. The estimation of the average uptake rate of total nitrogen in every treatment box

Parameter *)	Treatment box of α treatment						
Tarameter)	A	В	С	D	E	F	
Uptake rate by Lemna							
(mgTN/g/m²/day)	3.86	4.84	4.22	3.57	4.70	4.42	
Uptake rate by Lemna							
(mgTN/m²/day)	191.89	248.06	237.16	208.22	251.54	241.55	
Uptake rate by microalgae							
(mgTN/m²/day)	220.41	247.74	267.61	253.57	208.60	263.29	
D (*)	Treatment box β treatment						
Parameter *)	G	Н	I	J	K	L	
Uptake rate by Lemna							
$(mgTN/g/m^2/day)$	5.49	2.14	2.23	2.24	2.57	2.28	
Uptake rate by Lemna							
(mgTN/m²/day)	264.50	117.31	211.11	206.60	281.70	266.13	
Uptake rate by microalgae							
(mgTN/m ² /day)							

^{*)} in average wet weight

3.3. Uptake of Total Nitrogen pattern and profile

Figure 2 presents modelling on TN absorption (estimated based on the mass balance concept) results which show that microalgae community in treatment box of A, C, D, and I wereentirely superior in term of absorbing total nitrogen from its growth medium solution. Even cattapa extract (as bioalgaecide) was added in the box D, the TN absorbed by microalgae was still higher than that by the duckweed. Microalgae community also took more advantage when trace elementswereadded in TN uptake competition (treatment box of C and D) compares to the duckweed. It probably due to the dose of the bio-algaecide in this study inadequate to repress the microalgae growth. In treatment box of A, TN absorption exponentially started on the second day while the duckweedneeded longer time (on the sixth day). Both completed the exponential phase at day ten. In box C, D and I, both of microalgae and duckweedabsorbed TN immediately. It is suggested that the only NPK 100 mg/L added was not enough to facilitate faster TN absorption, but it must be enriched with trace element or doubling the NPK dose.

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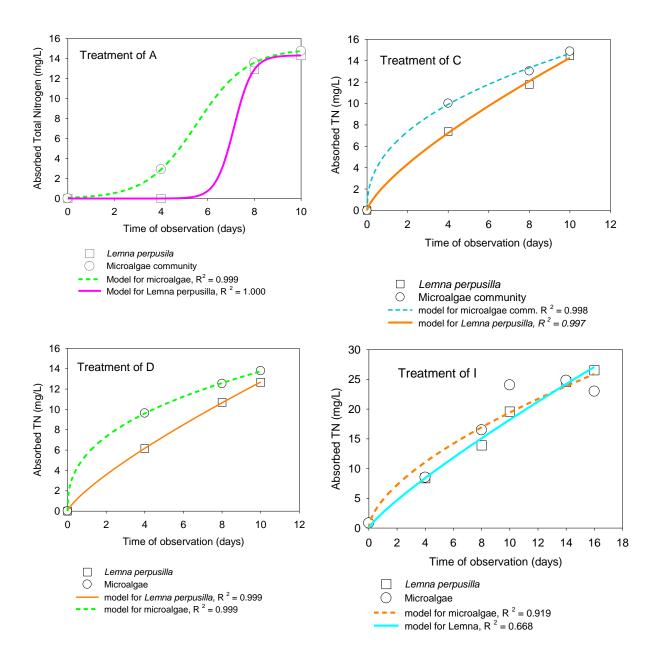


Figure 2. Total nitrogen absorption pattern which was *Lemna* out competed by microalgae during the Observation. Remarks: Treatment A: NPK100mg/L (control); Treatment C: NPK 100 mg/L+Trace Elements. Treatment D: NPK 100 mg/L +Trace Elements + Cattapa extract; Treatment I is NPK 200 mg/L.

Figure 3 presents the similarity pattern of TN absorbed by both of microalgae and duckweedwhich found in boxesF, G, and J. There was an alternate pattern in treatment of F, which initially the TN was more absorbed by the duckweedthan that of microalgae up to the second day, but outcompeted by the microalgae afterwards. On the contrary, in treatment G, the TN absorbed was faster by microalgae before day 3,which then took over bythe duckweeduntil day 8. The micoalgae took over back the lead on the TN absorptionat the same time, TN absorption in treatment J showed the opposite pattern of that the treatment G. These results demonstrate that even in box of F was enriched not only by trace element, molasses but also bio-algaecide, the duckweedstill failed to compete microalgae in terms of TN absortion. Instead, doubling the NPK dose combined with trace elementsenrichmentcan improve

16

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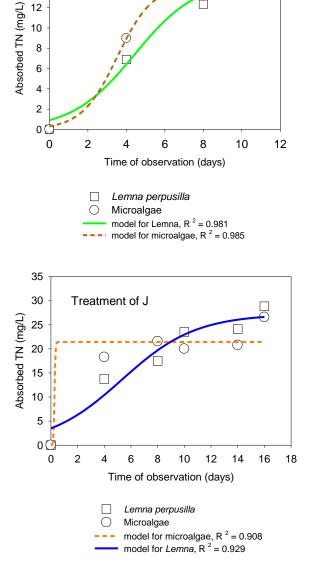
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Treatment of F

the duckweed performance in absorbing TN. It was indicated in box, when implied doubling NPK doses and adding trace element, an interesting competition pattern was occurred. Initially, that nutrient medium composition was more favorable for microalgae community in term of took up TN from medium solution. It proceeded faster than of duckweed biomass. Probably, it caused by the higher absorbing area on microalgae community than of duckweed biomass. The TN uptake by microalgae community then proceeded rapidly and led to quicker saturation (about one day after inoculation). On the contrary, the duckweed took more TN and gradually increased over the microalgae community (begun at the day of 9). It indicated that medium composition in the treatment box of J was the most optimum for the duckweed growth. A similar pattern also observed on duckweed growth experiment using palm oil mill effluent and waste water, therefore, it can be concluded that there was certain concentration of TN which is optimum for duckweed growth [18, 19]. In this study, that situation was observed on treatment box of J.



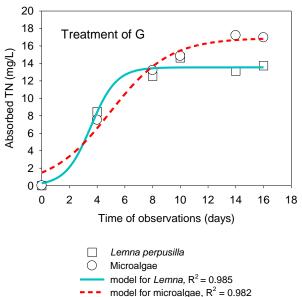


Figure 3. The alternately competition pattern between microalgae and Lemna for absorbing TN from its medium solution. Remarks: Treatment F (NPK mg/L+Trace Element+Molasses+Cattapa extract); Treatment G (NPK100mg/L); Treatment J (NPK 200 mg/L+ Trace Elements).

doi:10.1088/1755-1315/535/1/012027

Figure 4 presents the TN absorption profile of duckweed and microalgae in treatment box E. This box contained NPK 100 mg/L enriched with trace element and molasses. Curve fitting of this experiment data followed the sigmoid equation with a high degree of correlation which expressed as $R^2 = 0.999$. The duckweedabsorbed higher TN compared to microalgae from the beginning to the end of the experimentation time. Therefore, this curve proofs that the treatment E was the most preferred nutrient composition for culturing minute duckweed. Treatment of E also hadthe highest value in terms of TN uptake rate, which was as much as 251.54 mg/m²/day, remarkably higher than that of microalgae (208.60 mg/m²/day). It suggests that molasses in combined with trace elementsarea suitable nutrient formula to promoteduckweed optimal growth and simultaneously suppressed the microalgae growth.

Other phenomenonfound in this study is somesimilarity inTN uptake rate both of microalgae and duckweed (figure 4). Treatment B showed the highest similarity followed by treatment of K, both followed equation model of hyperbolic with two parameters. This box contained NPK of 100 mg/L mixed with bio-algaecide. Meanwhile, treatment of H and treatment of L were fitted with the model equation of sigmoid with three parameters. These results indicate that increasing NPK concentrations up to three folds resulted different TN absorption pattern when not in combined with trace elements solution (in case of treatment box K and L). The adding of bio-algaecide (B) and trace elements (H) also gave a different pattern of TN absorption, even mixed with NPK of 100 mg/L, which was formed hyperbolic response (faster) and sigmoidal response (slower).

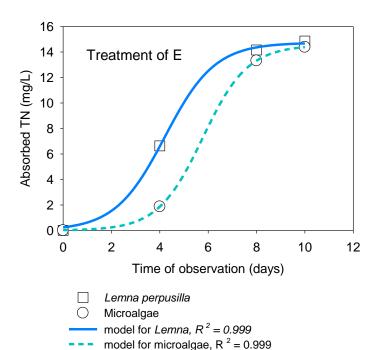


Figure 4. TN absorptionprofile of absorbed Total Nitrogen which shows TN Absorption of Duckweed higher than of microalgae. Remarks: Treatment of E was consisted of NPK 100mg/L+ Trace Elements +Molasses solutions.

doi:10.1088/1755-1315/535/1/012027

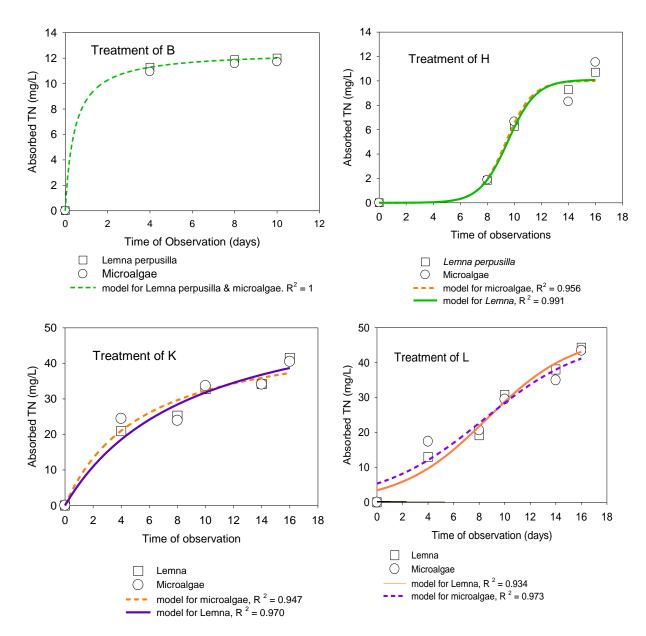


Figure 5. Similar pattern of TN absorption between of microalgae community and *Lemna perpusilla* in treatment box of B, H, K, and L; Remarks: Treatment B (NPK100 mg/L+ Cattapa extract); Treatment H Treatment K (NPK100 mg/L+ Trace Element); Treatment L (NPK 300 mg/L+ Trace Element).

Observation of chlorophyll-a content inthis study was carried out to represent the microalgae biomass development. Figure 6 describes the comparison between treatment medium A (as representative of the optimum condition for microalgal growth) and treatment medium E which was optimum for the growth of the duckweed. It is shown that chlorophyll-a content in treatment medium A was three folds higher than that of medium treatment E, which indicates the remarkable association between TN absorption with both the microalgal and duckweed biomass synthesis.

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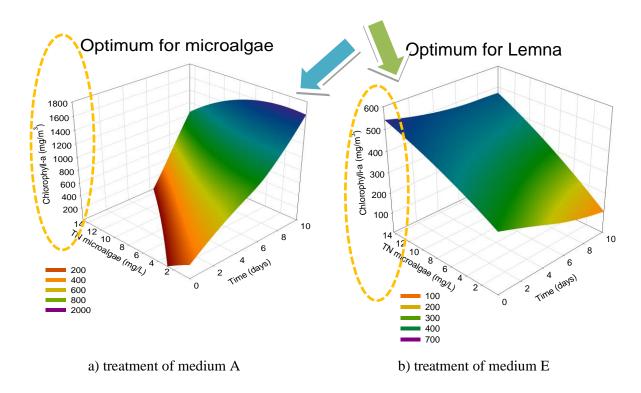


Figure 6. Time profiling of the chlorophyll-a content related to TN absorbed by microalgae.

The equation models and its determination coefficient (R^2) to describe the process which occurred in the 12 treatment nutrient composition are presented in Table 6. In general, the strength of correlation between experimental data and the model for treatment α (A to F) were higher than that of treatment β (G to L). The models for treatment I and J were not suitably fitted with the experimental data, which probably better to be simulated with other models. However, in case of other treatments (except of I and J treatments) models which presented in figures and equations are appropriate and would be useful for further implementation in the field, mainly for predicting as well as designing the desired processes.

doi:10.1088/1755-1315/535/1/012027

Table 6. Equation models on TN absorption in every treatment box, level of significant was held on α =0.05, passing normality distribution of Kolomogorov-Smirnov, p<0.001.

Box	Lemnaperpusilla as the	R ²	Microalgae community as the	R ²
	responding variable		responding variable	
Α	14.31	1.000	14.98	1.000
	$y_{Lemna} = \frac{1}{\left(1 + e^{-\left(\frac{x-7.33}{0.39}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x-5.53}{1.08}\right)}\right)}$	
В	12.53 <i>x</i>	1.000	12.53 <i>x</i>	1.000
	$y_{Lemna} = \frac{12.33x}{(0.44 + x)}$		$y_{microalgae} = \frac{12.53x}{(0.44 + x)}$	
С	$y_{Lemna} = 2.59x^{0.74}$	0.998	$y_{microglage} = 5.48x^{0.43}$	1.000
D	$y_{Lemna} = 2.07x^{0.78}$	1.000	$y_{microalgae} = 5.58x^{0.39}$	
Е	14 72	0.999	14 53	1.000
	$y_{Lemna} = \frac{117.2}{\left(1 + e^{-\left(\frac{x-4.21}{1.03}\right)}\right)}$		$y_{microalgae} = \frac{11.05}{\left(1 + e^{-\left(\frac{x - 5.78}{1.08}\right)}\right)}$	
F	14.76	0.981	14.25	0.985
	$y_{Lemna} = \frac{14.76}{\left(1 + e^{-\left(\frac{x - 4.43}{1.62}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x-3.52}{0.92}\right)}\right)}$	
G	13.54	0.984	16.91	0.982
	$y_{Lemna} = \frac{1000 \text{ m}}{\left(1 + e^{-\left(\frac{x-3.55}{0.91}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x-4.96}{2.12}\right)}\right)}$	
Н	10.09	0.956	10.01	0.956
	$y_{Lemna} = \frac{1}{\left(1 + e^{-\left(\frac{x-9.52}{1.04}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x-9.38}{0.98}\right)}\right)}$	
I	$y_{Lemna} = 2.61x^{0.84}$	0.668	$y_{microalgae} = 4.74x^{0.61}$	0.919
J	27.17	0.929	21.43	0.908
	$y_{Lemna} = \frac{1}{\left(1 + e^{-\left(\frac{x-5.32}{2.77}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x-0.27}{0.03}\right)}\right)}$	
K	60.6x	0.970	50.2 <i>x</i>	0.940
	$y_{Lemna} = \frac{1}{(9.07 + x)}$		$y_{microalgae} = \frac{1}{(5.54 + x)}$	
L	$y_{Lemna} = \frac{60.6x}{(9.07 + x)}$ 47.9	0.973	$y_{microalgae} = \frac{50.2x}{(5.54 + x)}$ 47.3	0.938
	$y_{Lemna} = \frac{1}{\left(1 + e^{-\left(\frac{X - 8.6}{3.37}\right)}\right)}$		$y_{microalgae} = \frac{1}{\left(1 + e^{-\left(\frac{x - 8.35}{4.06}\right)}\right)}$	

4. Conclusion

The Mass balance concept approach and modelling with non-linear equation was successfully employed to determine the most favorable medium composition in this study. The favorability is termed due to the medium capability to promote the minute duckweed growth while repressing the competitor microalgal growth. It proofed from the observation result which using 12 pre-formulated media. It had been revealed that medium E treatment (mixture of NPK of 100 mg/L enriched with trace element and molasses) was the most suitable medium composition. These results confirm the chlorophyll-a content which shows that NPK of 100 mg/L enriched with trace elements and molasses can be used for optimizing minute duckweed cultivation by repressing the microalgae community (as the growth competitor for duckweed).

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Acknowledgements

The authors wish to express their sincere thanks to the National Priority Program for Healing Lake Waters of RC for Limnology year 2019.