29. RICE STRAW FOR INHIBITING ALGAE AND THE EFFECTS OF PHENOL ON ALGAL GROWTH

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Harmful algal blooms have several impacts on human health, aquatic ecosystems and the economy. There are many ways to deal with algae problems including physical removal or poisoning. Rice is the major crop growth in Southeast Asia. If rice straw can use as algal control method then it is cheap, available and environmentally acceptable. This poster presents my ongoing research to use rice straw as an algaecide and determine chemical released during and after decomposing rice straw and its effects on algal growth.

Key Words : *Algal growth, Rice straw, Chlorophyll a, Phenol*

1. INTRODUCTION

Algae grow quickly related to algal bloom which caused by overabundance of nutrient and often bring about serious problems particularly in shallow eutrophic ecosystems. Some species produce toxins which have harmful effects not only on aquatic life but also on human beings. In addition, due to algal increase, bacteria use up the dissolved oxygen to decompose which causes the lack of oxygen in water.

In this study, we designed experiments and reported various factors such as Chlorophyll a (an indicator for algal biomass) DO, TN, NO_2-N, NO_3-N, TP, PO_4-P
Our objects are to determine chemical released during and after decomposing rice straw, the effect of rice straw and phenol on algal growth.

2. MATERIAL AND METHODS

(1) Chemical released during and after decomposing rice straw

To determine what chemical is released during and after decomposing rice straw, experiments was set by taking 5 *l* tap water and add 10 *g* rice straw which was cut into pieces as shown **Picture 1**, placed aeration and kept it in growth chamber at 35°C as shown **Picture 2**, light was set 24 hours.

(2) The effect of rice straw on algal growth

The water samples were taken containing

Picture 1 Dried rice straw 10g (2g/l)

Picture 2 Growth chamber

Microsystis areginosa and Microsystis wesenbergii which is one genus (Microsystis) forming heavy algal bloom in Japan. The water samples were collected from 10 to 20 *cm* from the water surface and stored at room temperature before experimenting. For details, it will be indicated in **part (4) The designed experiments**.

(3) The effect of phenol on algal growth

Algae water tanks M. aeruginosa (5*l*/tank) was collected from 10 to 20 *cm* from the water surface and stored at room temperature before experiment. Algae water mixed with 0.1*mg/l*, 0.05 *mg/l*, 0.005 *mg/l* phenol respectively .For details, it will be indicated in **part (4) The designed experiments**.

(4) The designed experiments

I designed 5 experiments including:

Experiment 1 (2013/10/1-2013/10/10): Rice straw was placed in tank which had only 5 *l* tap water and set up aeration. After that, tank was stored in growth chamber light is set 12 hours on and 12 hours off, 35°C (to hasten the rice straw decomposing process) Total Nitrogen, COD, Total Phosphorus, Phenol was measured.

To determine the effect of rice straw on algal growth, I performed 3 experiments (Experiment 2, experiment 3, experiment 4) by diluting water sample with distilled water until it reached a volume of 10 *l* divided in to 2 tanks wit 5 *l* each tank. Kept pH at 8.31, add 40 *ml* Media solution in each tank for culturing bloom-forming Microcysitis strains as shown in **Table 1**, store in in growth chamber at 35°C. Chlorophyll a, pH, TN, TN, NO_2-N, NO_3-N, TP, PO_4-P was measured.

Experiment 2 (2013/10/18-2013/10/22): Water sample contained Microsystis wessenbergii

Experiment 3 (2013/11/1-2013/11/14): Water sample contained Microsystis areginosa

Experiment 4 (2013/11/14-2013/11/17) Water sample contained Microsystis areginosa

For researching the effect of Phenol on algal growth, experiment 5 was set up.

Experiment 5 (2013/11/14-2013/11.19) Water sample contained Microsystis wessenbergii

Sample water which contains M. aeruginosa was mixed with with 0.1*mg/l*, 0.05 *mg/l*, 0.005 *mg/l* phenol respectively. Add 40 *ml* MA solution in each tank. Kept in growth chamber in 35°C, light was set 12 hours on and 12 hours off. Phenol, DO, Chlorophyll a was measured in experiment 5.

Table 1 Media solution

$Ca(NO_3)_2.4H_2O$	5 mg	$MnCl_2.4H_2O$	0.5 mg
KNO_3	10 mg	$ZnCl_2$	0.05 mg
$NaNO_3$	5 mg	$CoCl_2.6H_2O$	0.5 mg
Na_2SO_4	4 mg	$Na_2MnO_4.2H_2O$	0.08 mg
$MgCl_2.6H_2O$	5 mg	H_3BO_3	2 mg
$\beta - Na - glycerophosphate.5H_2O$	10 mg	Bicine	50 mg
$Na_2EDTA.2H_2O$	0.5 mg	Distilled water	100 ml
$FeCl_3.6H_2O$	0.05 mg	pH	8.6

3. RESULTS AND DISCUSSIONS

(1) Chemical released during and after decomposing rice straw

Total Nitrogen as shown in **Fig. 3a** , COD as shown in **Fig. 3b** Total Phosphorus as shown in **Fig. 3c** increase while Phenol as shown in **Fig. 3d** reduce. According to the result of experiment 1, there have appearance of amount of phenol tending to decrease with time. The degradation of phenol may be concern with high temperature and irradiation. In other hand, the appearance of heterotrophic bacteria can be a reason. Decomposing rice straw provide organic food source and where there is an organic food source, there will be heterotrophic bacteria. Heterotrophic bacteria include denitrifiers and sludge reducer (consume of organics), consume phenol. Since heterotrophic bacteria are facultative organisms, they can use either dissolved oxygen or nitrate as an oxygen source for metabolism and oxidation of organic matter. If dissolved oxygen and nitrate are present, bacteria will use the dissolved oxygen first. Experiment shows that TP and PO_4 increase. This problem maybe that bacteria are energy-limited rather than phosphate-limited so the amount of phosphate bacteria used is less than the amount of phosphate released.

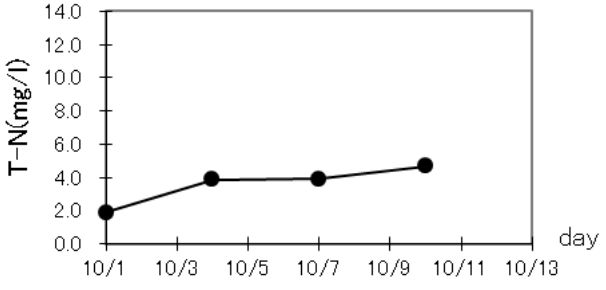


Fig. 3a Total Nitrogen in experiment 1

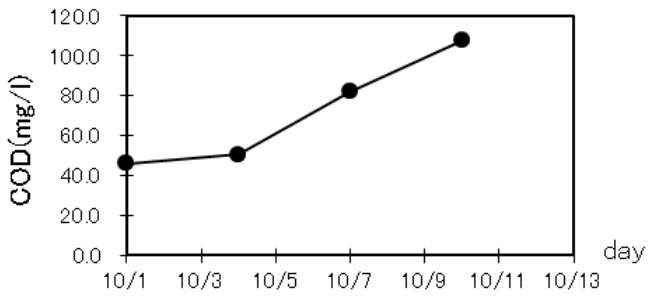


Fig. 3b COD in experiment 1

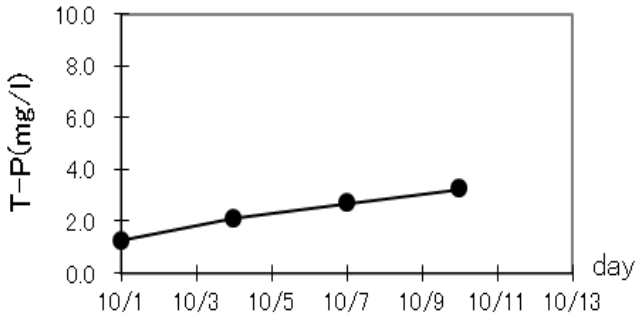


Fig. 3c Total Phosphorus in experiment 1

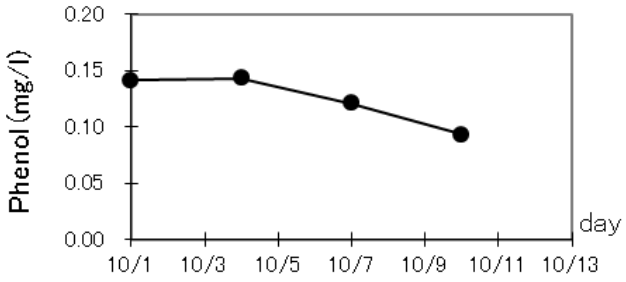


Fig. 3d. Phenol in experiment 1

(2) The effect of rice straw on algal growth

In experiment 2, Chlorophyll-a in case containing rice straw reduce quickly than case without rice straw as shown in **Fig. 4a** . pH of 2 case reduced, finally reached the same value as shown in **Fig. 4b**. The decrease of NO_2-N, NO_3-N as shown in **Table 2** in case using rice straw related to the decomposition of rice straw because of the lack of O_2 in water. Denitrification happen, heterotrophic bacteria get their oxygen by taking it off of nitrate molecules TKN increase as the increase of ammonium and pH being acidic.

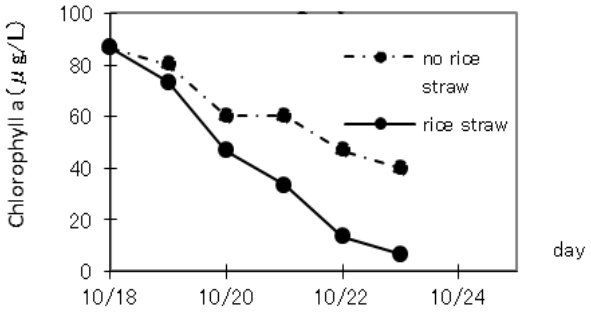


Fig. 4a Chlorophyll a in experiment 2

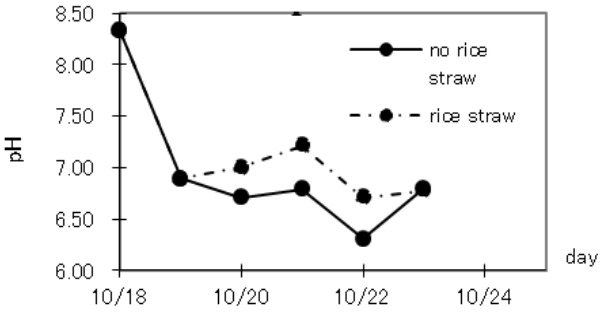


Fig. 4b pH in experiment 2

Table 2 TN, NO_2-N, NO_3-N, TP, PO_4-P during rice straw decomposing process in experiment2

No rice straw	T-N	NO2-N	NO3-N	T-P	PO4-P
2013/10/18	0.44	0.003	0.36	0.023	0.003
2013/10/19	-	-	0.42	-	-
2013/10/20	-	-	0.46	-	-
2013/10/21	-	-	0.21	-	-
2013/10/22	0.82	0.014	-	0.216	0.077
Rice straw	T-N	NO2-N	NO3-N	T-P	PO4-P
2013/10/18	0.44	0.003	0.36	0.023	0.003
2013/10/19	-	-	0.00	-	-
2013/10/20	-	-	0.00	-	-
2013/10/21	-	-	0.00	-	-
2013/10/22	2.20	0.001	-	0.597	0.653

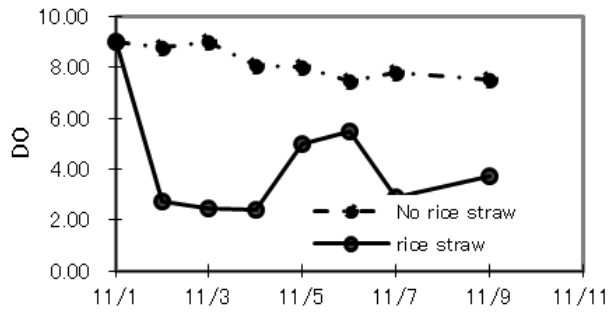


Fig. 5a DO in experiment 3

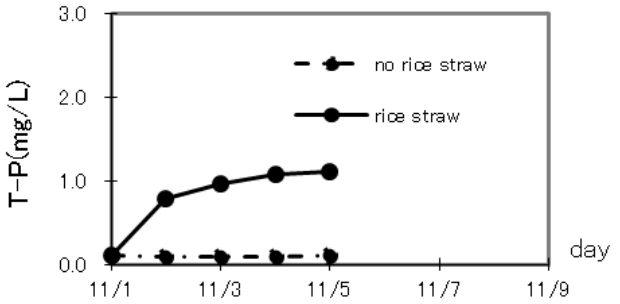


Fig. 5b TP in experiment 3

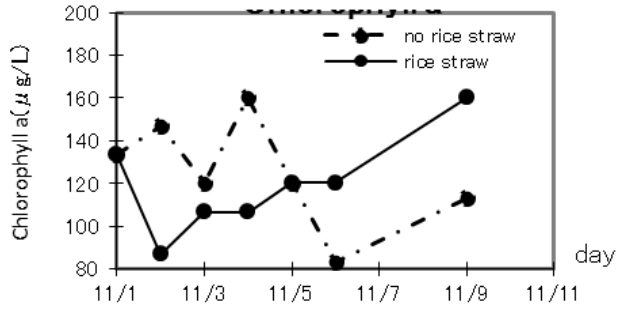


Fig. 5c Chlorophyll a in experiment 3

Table 3 pH., turbidity, T-N, NO_2-N, NO_3-N in experiment 3

No rice straw	pH	Turbidity	T-N	NO2-N	NO3-N
2013/11/1	8.31	4.6	1.49	0.012	0.24
2013/11/2	7.98	8.6	1.20	0.018	0.20
2013/11/3	8.31	9.8	1.36	0.007	0.00
2013/11/4	8.52	11.3	1.31	0.001	0.00
2013/11/5	8.31	9.8	1.33	0.002	0.00
Rice straw	pH	Turbidity	T-N	NO2-N	NO3-N
2013/11/1	8.31	4.6	1.49	0.012	0.24
2013/11/2	7.22	18.2	2.40	0.004	0.00
2013/11/3	7.32	22.1	2.34	0.003	0.00
2013/11/4	7.12	16.0	2.17	0.003	0.00
2013/11/5	7.66	15.6	2.31	0.004	0.00

In experiment 3, Phosphate in case using rice straw is high as the result of phosphorus released by rice straw decomposition and bacteria decomposition converts organic phosphorus in dead algae back to inorganic phosphorus DO in case without rice straw is higher than case containing rice straw as shown in **Fig. 5a**. Chlorophyll a in case containing rice straw went down quickly from 11/1 to 11/2 than recover quickly and higher than the mount in case without rice straw as shown in **Fig. 5c**. TN, NO_2, NO_3, NH_4, TP, PO_4 of case using rice straw is higher than those indication of case without rice straw as shown in **Fig. 5b** and **Table 3**

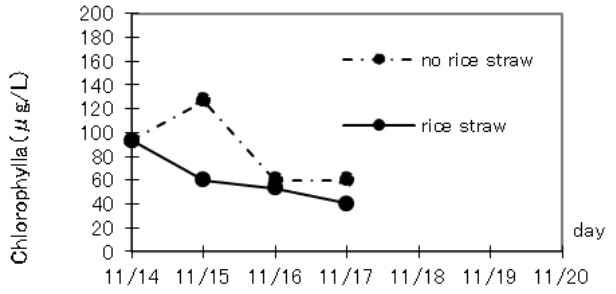


Fig. 6b Chlorophyll a in experiment 4

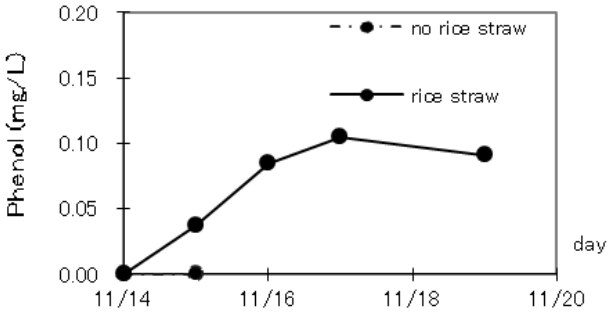


Fig. 6c Phenol in experiment 4

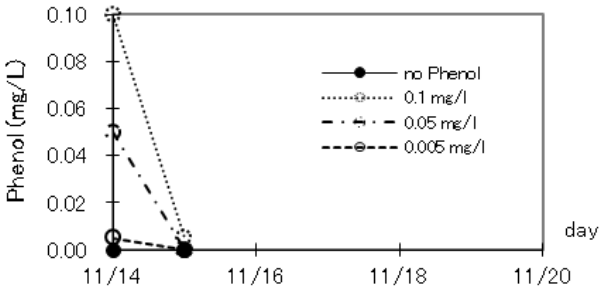


Fig. 7a Phenol in experiment 5

In experiment 4, the suppression of DO relates to the decomposition of organic matter.

Phosphorus or Nitrogen is not the main reason of Chlorophyll-a recover because Phosphorus and Nitrogen emission during process.

The pH and Chlorophyll of case without rice straw was higher than case using rice straw as shown in **Fig. 6a** and **Fig. 6b**. Phenol also reduce gradually as shown in **Fig. 6c**

The suppression of DO related to the decomposition of organic matter when rice straw decomposition.

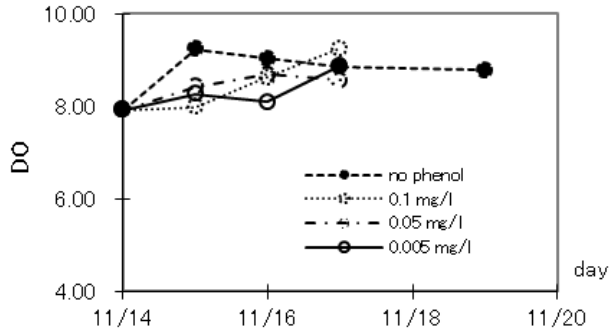


Fig. 7b DO in experiment 5

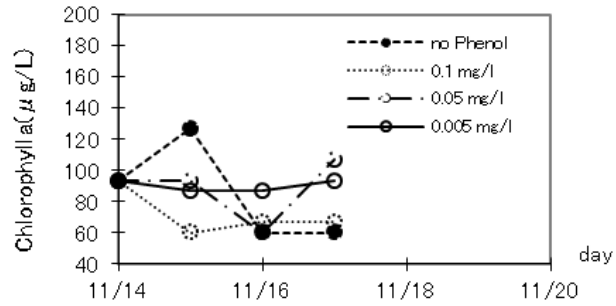
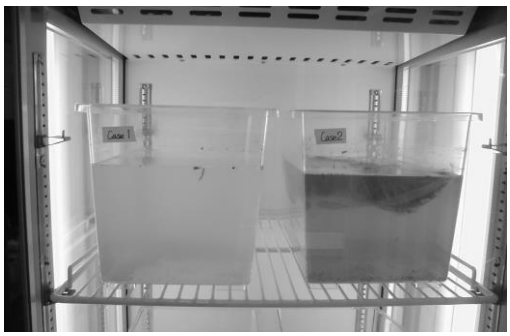


Fig. 7c Chlorophyll-a in experiment5

Picture 3 Water sample (2013/11/14)



Picture 4 Water sample (2013/11/19)

(3) The effect of Phenol on algal growth

Phenol with designed dose disappeared quickly as shown in **Fig. 7a**. DO in case using Phenol tend to increase from 11/17 as shown in **Fig. 7b**. The amount of Chlorophyll a in case using rice straw reduced at start and recover and grow quickly as shown in **Fig. 7c**

Based on experimental results, Phenol can inhibit the algal growth. Phenol dose is also important for algae inhibition. However Phenol disappears quickly and then algae recover quickly.Due to the decomposition of rice straw, organic matter decomposition effect on bodies of water. It make water to turn brown as shown in **Picture 3** and **Picture 4.**

4. CONCLUSIONS

This study shows that it is not convincing to conclude that straw provides carbon source for carbon-limited microbial growth and as the microbes grew, they use phosphorus. Algal growth would be limited by a lack of phosphorus (Steve Mccomas et al 2008, Anhorn et al 2005) Base on experiments Phenol is a main reason for algal inhibition not Phosphorus dose.

Using rice straw means providing organic food source for heterotrophic bacteria. In addition, there are many species of heterotrophic bacteria are facultative anaerobes, they can function with or without oxygen. It will compete with autrophic bacteria of oxygen and surface area and they can reduce oxygen levels related to "bacteria bloom"

5. REFERENCES

1) Effect of light, temperature, and pH on the accumulation of phenol by Selenastrum capricornutum, agreen alga, *John L. Newsted*
2) *Ichimura*, T.1979 2. Isolation and culture methods of algae 2.5.B. Freshwater algae
3) Barley straw installation and water quality conditionsin Valley Lake, Lakeville, Minnesota, 2008, *S. McComas and J.Stuckert, January 2009*
4) Effect of light, temperature, and pH on the accumulation of phenol by Selenastrum capricornutum, agreen alga, *John L. Newsted*
5) Photosynthetic nitrate assimilation in cyanobacteria, *Enrique Flores, Jose E. Frias 2004*
6) Rethinking the role of Nitrogen and Phosphorus in the Eutrophication of aquatic ecosystems, *Karl Havens, Thomas Frazer 2013*