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# 4 SPIRULINA PRODUCTION IN FERTILIZER MEDIUM COMBINED BY TOFU AND LATEX LIQUID WASTES

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## Abstract

Spirulina is one of microalgae that used in nutraceutical dan pharmaceutical industries. The biomass could produced in liquid waste of tofu and latex industries. The aims of this research were to know the influence tofu and latex liquid waste percentage in medium of cultivation for their maximum density and specific growth rate. This research was arranged in a factorial groups randomized design with two treatment factors (kinds of liquid wastes and their percentage in Spirulina medium) and three groups based on light intensities. Spirulina production could used tofu and latex liquid wastes. One hundred percent tofu liquid waste gave highest density and 75% tofu liquid wastes gave maximum specific growth rate as like as 100% fertilizer for Spirulina medium. Light intensity 1000 – 4000 lux could be seen on the Spirulina cultivation if they were cultured in fertilizer medium was combined by tofu and latex liquid wastes.

## Keywords

Spirulina, tofu, latex, liquid waste

## INTRODUCTION

*Spirulina platensis* is a prokaryotic organism which contains a lot of bioactive compound in their biomass (Henrikson, 1989). Filamentous microalga *S. platensis* (cyanobacteria) has been produced commercially all over the world due to its high content of protein (up to 70%), pigments (especially the blue pigment phycocyanin), essential fatty acids (e.g.,  $\gamma$ -linolenic acid), vitamin B12 and minerals (Goksan, Zekerruyaolulu, 2007). In addition, it is successfully used in aquaculture and poultry industries as well. *Spirulina platensis* have been used for cosmetic, food, nutraceutical and pharmaceutical industry. *Spirulina platensis* has been cultured in serum latex medium that skim effluent from latex industry, and used for reducing COD, total nitrogen, and ammonia nitrogen concentration of the medium optimally (Tri Panji *et al.*, 1995; El-Baky *et al.*, 2008). The microalgae cells that be cultured in this effluent can be bioremediator and a source of some bioactive compound for material industry.

*Spirulina platensis* is the most widely studied photosynthetic microorganism, only a few attempts have been made up to now to utilize it for removal of nutrients from wastewaters (Converti *et al.*, 2006). *Spirulina* biomass can cultivated in organic liquid waste such as tofu and latex industry. Tofu and latex liquid wastes have organic

matter which can be used for heterotroph cultivation of *Spirulina*. Composition of liquid waste in *Spirulina* medium needed optimizing for biomass production. The aims of this research were to know the influence tofu and latex liquid waste percentage in medium of cultivation for their maximum density and specific growth rate.

## MATERIALS AND METHODS

*Spirulina platensis* were obtained from Indonesian Biotechnology Research Institute for Estate Crops (Bogor, Indonesia). They was cultured in modified *Spirulina*'s technical medium (Wijayanti, 1999) during 60 days at Aquaculture Laboratory. Technical medium contain  $MgSO_4$  0,2 g.l<sup>-1</sup>,  $CaCl_2$  0,04 g.l<sup>-1</sup>, EDTA 0,08 g.l<sup>-1</sup>, TSP 0,5 g.l<sup>-1</sup>, Urea 0,3 g.l<sup>-1</sup>, ZA 1,32 g.l<sup>-1</sup>,  $NaHCO_3$  8,5 g.l<sup>-1</sup>, A<sub>5</sub> solution 0,1% v/v. Biomass was isolated in batch and non axenic system.

This research used tofu and latex liquid wastes from effluent waste water of tofu and latex industry at Palembang and Ogan Ilir, South Sumatra. The liquid wastes were prepared with 100°C boiling and filtering (plankton net mesh size 25µm). Liquid wastes and isolated composition were arranged in a factorial group randomized design with two treatment factors, kinds of liquid wastes (tofu and latex liquid medium) and their percentage in *Spirulina* medium (0, 25, 50,75,100% liquid waste in cultivation medium) and three groups based on light intensities consisted of low intensity 500-2500 lux, medium intensity 1000- 4000 lux and high intensity 2000-6000 lux approximately.

Density of biomass measured with Genesys-20 spectrophotometre in 560 nm wavelength at 0,1,2,3,7,14,21,25,26,27, 28 days cultivation. The absorbance was converted in gram of dry biomass per litre of cultivation medium. Specific growth rates ( $\mu$ ) were calculated as following equation (Becker,1996) :

$$\ln N_t - \ln N_0 = \mu \cdot t + C$$

Where  $N_0$  is density of *Spirulina* biomass (g.l<sup>-1</sup>) at zero time,  $N_t$  is density of *Spirulina* biomass (g.l<sup>-1</sup>) at t days cultivation, t is time of days cultivation. Datas were calculated in logarithmic phase of growth.

Datas were analyzed by ANOVA (Analysis of Variance) and were followed with LSD (Least Significance Difference) test if f test showed difference number significantly

## RESULT AND DISCUSSION

Table 1 shows maximum density of *Spirulina* biomass ( $\text{g.l}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium. Maximum density of *Spirulina* biomass which cultivates in tofu liquid waste was higher ( $3,36 \text{ g.l}^{-1}$ ) than in latex liquid waste medium ( $2,70 \text{ g.l}^{-1}$ ). The nutrients from tofu liquid wastes were more adequate for *Spirulina* growth than from latex liquid waste. Composition 50% and 75% of liquid wastes in *Spirulina* medium gave highest average density of *Spirulina* biomass  $2,97$  and  $3,23 \text{ g.l}^{-1}$ . Interaction of treatment 50, 75, 100% tofu liquid wastes and control medium gave highest density of *Spirulina* between  $3,48 - 3,89 \text{ g.l}^{-1}$ .

Table 1. Maximum density of *Spirulina* biomass ( $\text{g.l}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium.

Composition of liquid waste percentage	Kind of liquid waste influence		Percentage of waste influence
	Tofu	Latex	
0%	3,89 e	3,89 e	3,89 c
25%	2,06 ab	2,88 bcd	2,47 a
50%	3,48 cde	2,46 b	2,97 ab
75%	3,67 de	2,79 bc	3,23 b
100%	3,68 de	1,49 a	2,58 a
Kind of liquid waste influence	3,36 b	2,70 a	

Table 2 shows average of maximum density ( $\text{g.l}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium at three groups based on light intensity. Heterotrophical cultivation of *Spirulina* grew optimally in medium of light intensity between 1000-4000 lux approximately. The average density of *Spirulina* biomass which cultivated with medium lightness was highest ( $3,44 \text{ g.l}^{-1}$ ) as like as cultivation with 4h lightness.

Table 2. Maximum density of *Spirulina* biomass ( $\text{g.l}^{-1}$ ) at three groups based on light intensity.

Group of light intensity	Average maximum density	LSD (0,05)
High	3,06	ab
Medium	3,44	b
Low	2,59	a

Specific growth rate of *Spirulina* logarithmic growth phase showed adaptability in their cultivation medium. Table 3 shows specific growth rate of *Spirulina* biomass ( $\%.\text{day}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium. Composition 0% waste gave highest of specific growth rate ( $16,58 \%.\text{day}^{-1}$ ). It is possible because they were cultivated in the same of medium when they were prepared before this research. In liquid waste, 50 and 70% composition were the best of liquid waste medium for the specific growth rate. Tofu liquid waste gave influence better than latex liquid wastes. Nutrient from tofu liquid waste

could be easier to be adsorbed by *Spirulina* cell than nutrient from latex waste. Interaction of kind and percentage of liquid wastes showed 50% and 75% tofu liquid wastes were not different with technical medium significantly.

Table 3. Specific growth rate of *Spirulina* biomass ( $\%.\text{day}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium.

Composition of liquid waste percentage	Kind of liquid waste influence		Percentage of waste influence
	Tofu	Latex	
0%	16,58 d	16,58 d	16,58 c
25%	7,29 a	10,54 bc	8,92 a
50%	13,57 cd	9,08 ab	11,33 ab
75%	13,50 cd	10,97 bc	12,24 b
100%	11,47 bc	5,81 a	8,64 a
Kind of liquid waste influence	12,48 b	10,60 a	

Table 4. Specific growth rate of *Spirulina* biomass ( $\%.\text{day}^{-1}$ ) at three groups based on light intensity.

Group of light intensity	Average maximum density	LSD (0,05)
High	11,62	ab
Medium	13,20	b
Low	9,81	a

Table 4 shows average of specific growth rate ( $\%.\text{day}^{-1}$ ) which were cultured in tofu and latex liquid wastes combined *Spirulina*'s technical medium at three groups based on light intensity. In medium and high lightness (approximately 1000-6000 lux), heterotroph cultivation was more suitable for degradation of organic compound to be an inorganic compound which was done by symbiotic microbe in the medium.

Liquid wastes can contain metal ions, included heavy metals (Pb, Hg, Cd) that are hazardous for human consumption such as food, nutraceutical and pharmaceutical. Cyanophyta such as *Spirulina* have capability for metal adsorption 88-97% (da Costa and de Franca, 2003; Inthorn, *et al.*, 2002). Adsorptive capacity is influenced by many factors including: properties of metal ions in aqueous solution, biosorption conditions (such as pH, temperature, contact time, the presence of other ions in the solution, initial concentration of metal ions and the biomass) and algal species (Michalak *et al.*, 2007). Pretreatment of liquid wastes is needed for safety. Heavy metal removal can be done by precipitation, filtration or growing aquatic plant for adsorption of metal ions and removal nitrogen and phosphorus exceed in the organic liquid wastes. *Spirulina* have limiting factor for their growth. One of the limiting factors is maximum nitrogen concentration. Maximum nitrogen concentration able to sustain the batch growth of this microalga without inhibition was 23,8 ppm (Converti *et al.*, 2006). Tofu and latex liquid wastes contains exceed nitrogen until 250 ppm (Wi-

jayanti and Jubaedah, 2009), so that is needed to remove nitrogen for optimizing Spirulina growth.

## CONCLUSSION

One hundred percent tofu liquid waste gave optimum density and 75% tofu liquid wastes gave maximum specific growth rate as like as 100% fertilizer for Spirulina medium. Light intensity 1000 – 4000 lux could be given on the Spirulina cultivation if they were cultured in fertilizer medium was combined by tofu and latex liquid wastes.

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