Improving the Quality of "*Leri*" Rice Washing Waste by Different Period of Fermentation and Yeast Concentration as an Alternative Liquid Organic Fertilizer

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Abstract: This study aims to determine the best time of fermentation process and yeast concentration to improve the quality of "leri" (rice wahing waste). The study was conducted in the Bilibili village, Suppa Sub-District, Pinrang Regency, South Sulawesi Province from April to July 2012. Samples were analyzed at the Laboratory of Chemical and Food Livestock, Hasanuddin University, and Laboratory of Chemistry Department of Mines and Energy, Makassar. The study used a Completely Randomized Design (CRD) factorial pattern. The first factor i.e., treatment of fermentation period consist of a control, 2, 4, and 6 of fermentation period. The second treatment is the mass of yeast consisting of a control 1, 2, and 3 g of yeast per 5 liters of leri. Data were analyzed by two factor analysis of variance without replication. Duncan test was used for significant treatment. The result shows that the time of fermentation for 6 days produce relatively high ethanol (0,52 %), increasing the mineral content of phosphorus (0,15 ppm) and sulfur (0,35 %), and mineral content of nitrogen are relatively good (0,11 %). Yeast 3g per 5 liters of leri, gave betterethanol result (0,43 %), increased mineral content of nitrogen (0,11 %) and phosphorus (0,16 ppm), and potassium mineral content were quite good (350,25 ppm).

Keywords: Leri waste; fermentation period; liquid organic fertilizer; yeast; Saccharomyces

1. Introduction

Alternative crop fertilizer materials have been used and proven empirically from generation to generation to cause plants to flourish. One of the materials that can be used is the waste water from washing rice, which is also called "leri" (Java language). Leri can be used to improve plant growth, especially at first washing leaving thick white solution which contain carbohydrates (starch), gluten, cellulose, hemicelluloses, protein, thiamin (Vitamin B1), Vitamin B12, P and Fe minerals.

Leri has been studied in several researches, such as for breeding media of *Bacillus thuringiensis* local strains H-14 (Yuniarti and Blondine 2008), nata de leri production (Bambang, 2009); Layiatul, 2010) making sirup (Aminah *et al.*, 2013), alternative media carrier for *Pseudomonas fluorescence*, where the bacteria are microbes that play a role in controlling of rust disease-causing pathogens and trigger the growth of plants (Nurhasanah *et al*., 2010). On the other hand, research on utilization of leri as fertilizer has been conducted on many plant species such as *Lactuca sativa* plants (Wulandari, *et al*, 2011), Adenium (Andrianto, 2007), on tomato and eggplants (Arin, 2008 and Leandro, 2009 *in* Istiqomah (2012), respectivel, *Pleurotus ostreatus* (Kalsum *et al.*, 2011), and Istiqomah (2012) on green bean plants, while Ni Luh (2014) on orchid. Species. However the utilization of waste leri mentioed here were given directly without any increase in effluent quality. Leri quality improvement can be made through fermentation technology by utilizing microorganisms such as *Rhizopus, Aspergillus, Mucor, Amylomyces, Endomycopsis, Saccharomyces, Hansenula anomala,, Lactobacillus, Acetobacter*, etc. which are are present in yeast, to breakdown sugar carbohydrates contained in leri into bioethanol.

Yeast has been widely used commercially to produce ethanol than bacteria and fungi, this was due to the yeast which can produce ethanol in large quantities and have tolerance to high ethanol levels. At optimum condition ethanol concentration may be produced 8-20%. Yeast can change liquid containing sugars into ethanol and CO_2 gas quickly and efficiently.

Ethanol in low concentration itself has been shown to affect growth of plants. It is thought that somehow it may affect growth of by triggering available CO_2 for photosynthesis. The result of research Akib and Gusri (2008) using different concentration of ethanol, conclude that for plant growth analysis parameters leaf area index (LAI) and net assimilation rate (NAR) of soybean (*Glicena max*) were best obtained on the application of ethanol with a concentration of 10% and 20%. Siro (1965) in Akhsan and Gusri (2007), reported that the optimal ethanol concentration tested for flower formation in *Nicotiana tabacum*, was not detected in the range of concentrations used. Kazumitsu and Sato (1996) in Akhsan and Gusri (2007) reported that ethanol treatment, can break dormancy rice seed rice of both japonica and indica varieties. Based on these descriptions, this study try to prove the hypothesis by improving quality of leri. What is the concentration of yeast that will enhance the quality of leri and how much

fermentation time should be answered.

2. Materials and Methods

2.1 Research Methodology

The study used Completely Randomized experimental design (CRD) in two factors factorial pattern. First factor was treatment of fermentation period which consisted of a control 2, 4, and 6 days of fermentation. The second treatment was the concentration of yeast consisting of a control, 1, 2, and 3 g per 5 liters of leri. The leri used was of first rice washing, obtained from the liquid waste generated by restaurants and food stalls, which further homogenized and sterilized before, so the rice varieties, the amount of water and volume of rice used that is not a factor to consider. Each treatment was combined in order to obtain 16 combinations of treatments, which is repeated three replications so that there are 48 experimental units. For the chemical analysis purposes (ethanol, minerals of nitrogen, phosphorus, potassium, and sulphur content) a composite sample of three replicates for each treatment were made per parameter. Chemical Analysis for nitrogen, potasium and sulphur were conducted in Laboratory of Food Chemistry and Livestock University of Hasanuddin, while analysis of ethanol content and mineral of phospor were conducted in Laboratory of Chemistry Department Mines and Energy, Makassar.

2.2 Data Analysis

Data observed were analyzed with factorial pattern without replication which only use data from the interaction between both treatments to see the effect of each treatment. Duncan test were performed on treatment showing significant effect (Akib, 2013).

3. Results and Discussion

3.1 Content of Ethanol

The results of analysis of variance on fermentation time and yeast concentration, are shown a significant effect for the content of ethanol. The average value of treatment and Duncan test results can be seen in **Figure 1**.

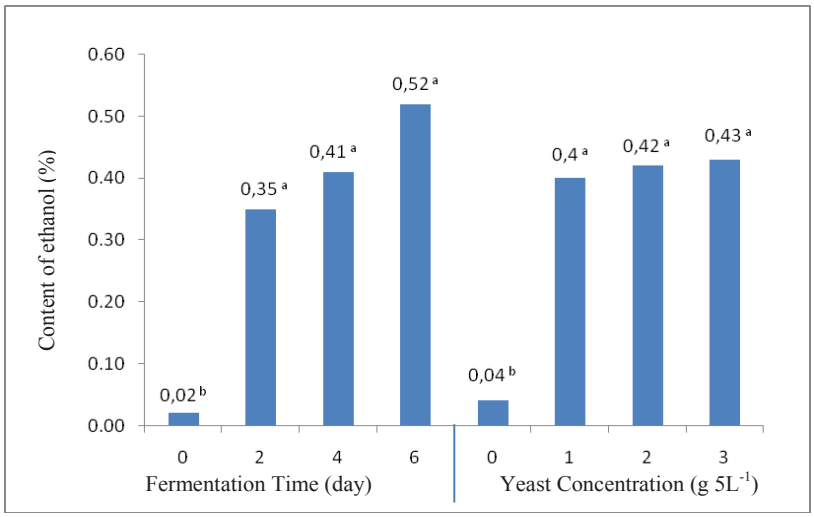

Figure 1. The average ethanol content at treatment in different fermentation time and yeast concentration. The numbers followed by same letters are, not different significantly based on Duncan test at level $\alpha = 0.01$.

Figure 1 illustrated that the longer leri is fermented, the content of ethanol produced is higher, which was predictable sincet the time of fermentation given as treatment, provided enough time to microorganisms (*Rhizopus, Aspergillus, Mucor, Amylomyces, Endomycopsis, Saccharomyces, Hansenula anomala, Lactobacillus, Acetobacter, etc*.) contained in the yeast to break down the sugar contained in the leri, which reaching 58-90% (Nurhasanah, *et al*., 2010). Ajay *et al.* (2014) concluded in his study that the change in the concentration of yeast, the time required for the completion of fermentation decreased. The longer fermentation process would provide opportunities for enzymes to break down the sugar into alcohol (Setyohadi (1993) in Sebayang, 2006; Aarti and Anita, 2010). The presence of high concentrations of yeast meant increasing number of yeast cells which was followed by an increase of enzymeSs produced to break down sugars into ethanol (Deky, *et al*, 2012). The more an enzyme was produced the sugar conversion process by enzyme in to alcohol became increasingly rapid (Judoamidjojo, 1990). Catabolism of glucose to ethanol by yeast was an attempt to obtain the necessary energy in growth. So the amount of ethanol produced also depends on the available sugar (Firman, 2006) or C/N ratio (Anggraeni *et al*,. 2010) in the substrate. Supriyanto (1995) and Kusnadi *et al*., (2009), also concluded that the period of 6 days fermentation could have resulted in the highest content of ethanol. After day 6, content of ethanol will drop because the process of fermentation continued from ethanol to acetic acid.

3.2 Mineral of Nitrogen

Analysis of variance showed that fermentation time treatment and yeast concentration had noaffect to nitrogen mineral content. The highest content of mineral nitrogen was obtained on the treatment of 4 days fermentation period and yeast concentration of 3g per 5 liters of leri (**Figure 2**).

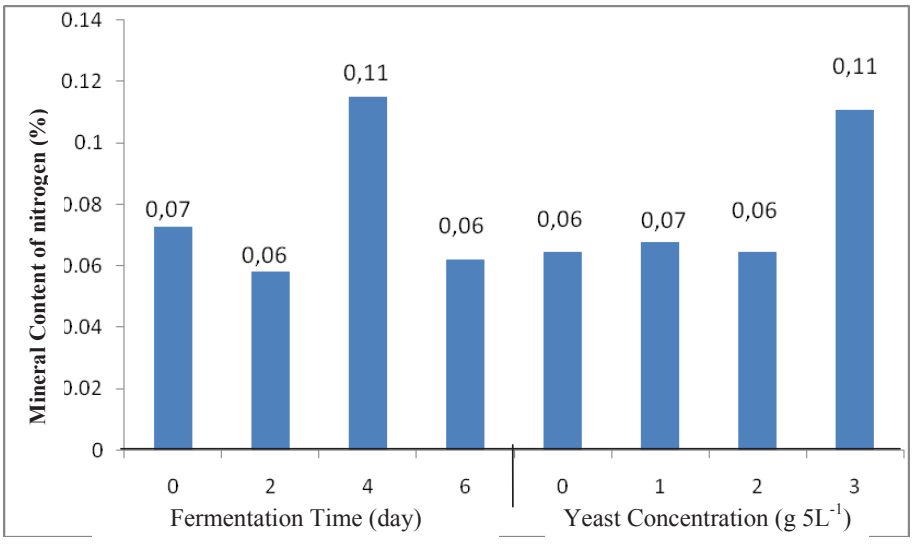

Figure 2. The average mineral content of nitrogen at treatment in different time fermentation and yeast mass.

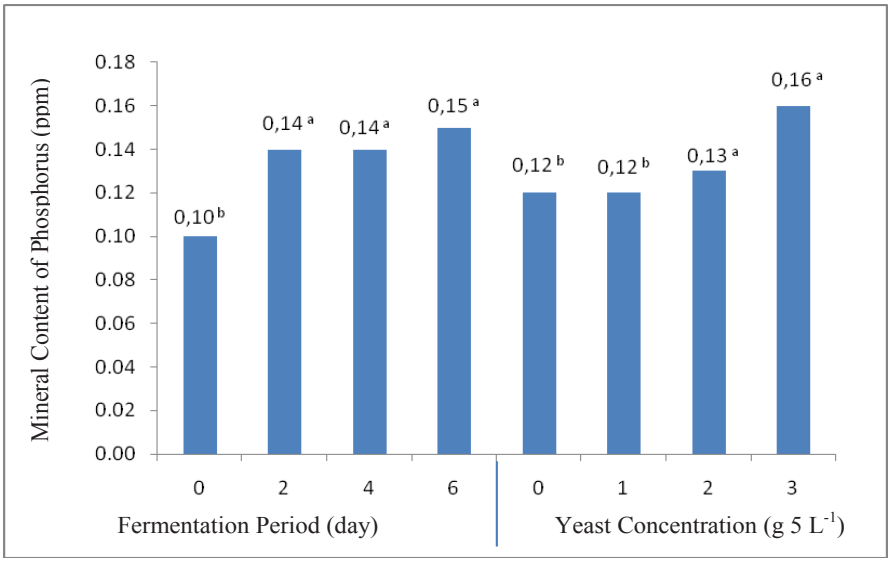

Figure 3. The average yield of mineral content of phosphorus after treatment of different fermentation duration and yeast concentration. The numbers followed by same letter, are not different significantly based of Duncan test at level α = 0.01.

It was assumed that on day 4 of the fermentation period with yeast concentration of 3 g per 5 liters of leri, the microorganism worked on its maximum and producing ATP (Adenosine Tri Phosphate), which is an nucleotide containing mineral nitrogen (Yudiarto, 2008). The content of mineral nitrogen on ermentation for 6 days has decreased, this case was thought to be caused by microorganisms dying out because nutrients availablility had expired. Microorganisms required nutrients for synthesis of cellular components to produce energy. This is consistent with results found by Fardiaz (1989) which concluded that population of micro-organisms begin to die as a result of nutrients in medium and energy reserves in cells had been discharged. Furthermore, Fardiaz (1989) explained that nutrient content of mineral nitrogen, is third highest after carbon and oxygen mineral. Function of nitrogen mineral in microbial cell physiology is part of a protein, nucleic acid, and co-enzyme.

3.3 Mineral of Phosphorus

The results of statistical analysis showed that duration of fermentation period and yeast concentration (6 day and 3 g per 5 liters of leri), significantly to mineral content of phosphorus. The highest mineral content of phosphorus was obtained in treatment fermentation for 6 days and yeast concentration of 3 g per 5 liters of leri. The results of average values and Duncan test can be seen in **Figure 3**.

During fermentation period of 2 and 4 days the microorganisms was just beginning the process of adaptation and had not yet working optimally. and after 6 days the microorganisms have enough time to break down maximally the sugars to produce energy with the nutrients available. Separately, at yeast concentration of 3 g per 5 liters of leri had increasedd the mineral content of phosphorus, which may be due to the inreased number of microbial cellsbreak down carbohydrates. So the phosphor mineral content become nutrients in the

process of fermentation. Chuzaemi (1994) in Suprayogi. 2010, reported that the microbial production is limited by the availability of energy, soluble protein, and minerals especially of phosphorus.

3.4 Mineral of Sulfur

The function of sulfur minerals in the physiology of microbial cells are part of protein (amino acids cysteine and methionine) and part of several enzymes (CoA, Co-enzyme A ocarboxsylase).

The analysis of variance, indicated that treatment of fermentation period and yeast concentration had no significant effect on the yield of mineral content of sulfur. Average of mineral content of sulfur was highest obtained at treatment of 6 days of fermentation period and without addition of yeast mass (control) as shown in **Figure 4**. The large mineral content of sulfur on the treatment of 6 days fermentation, were assumed as a result from other sulphur minerals derived from *Saccharomyces cerevisiae* (the yeast) itself, *Saccharomyces cerevisiae* also have more time to produce enzyme, so that mineral content of sulfur on treatment of fermentation period of 6 days is relatively higher. On the other hand, at the treatment of yeast concentration, mineral content of sulfur was obtained relatively higher on the control treatment. We assumed that the mineral content of sulfur contained in the leri was not consumed by *Saccharomyces cerevisiae* to survive by breaking down sugars into ethanol. Prescott and Dunn (1959) in Maggy (1990) stated that, other than source of carbon, *S. cerevisiae* also required a source of sulphur minerals and vitamins in its growth. The results of the study of Widayanti *et al*. (2013) proved that the addition of mineral sources of sulfur in the media, showed that sulphur was not a source of nutrients to *S cerevisiae* to produce alcohol.

3.5 Mineral of Potassium

The results of analysis of variance showed that treatment of fermentation time had significant effect on potassium content

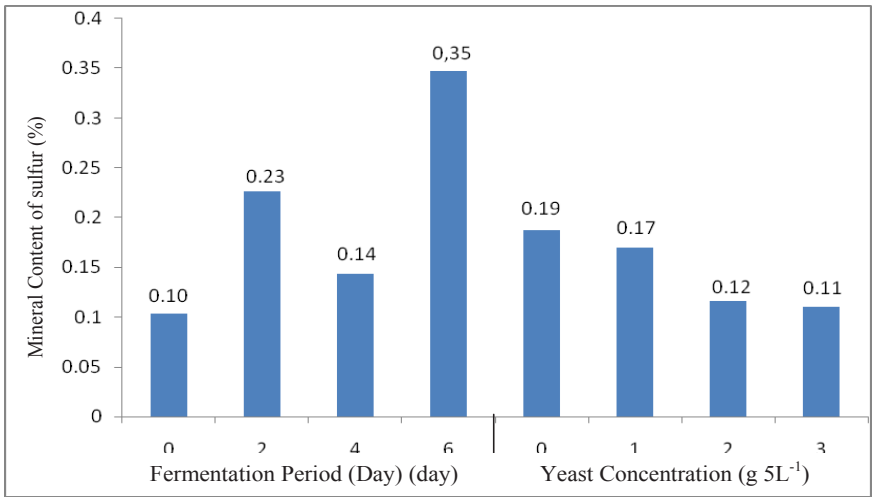

Figure 4. The average of mineral content yield of sulfur at different fermentation period and yeast concentration.

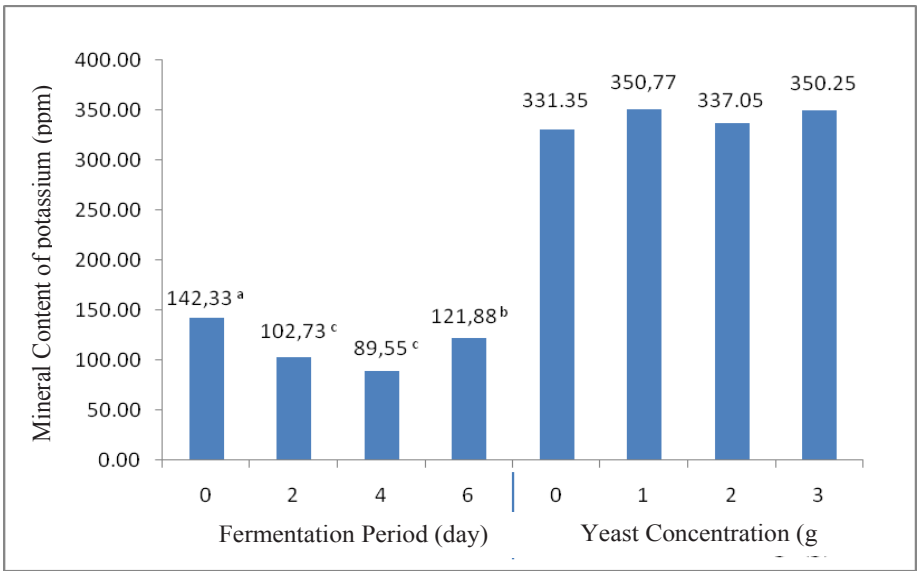

Figure 5. The average yield of mineral content of potassium at treatment of different time fermentation and yeast concentration. The numbers followed by same letters were not different significantly based of Duncan test at level $\alpha = 0.01$.

but was not significant at concentration treatment of yeast. The average yield of mineral content of potassium was obtained on treatment without fermentation (control). Whereas for treatment of yeast concentration, the mineral content of potassium highest was obtained at treatment of mass yeast 1 g per 5 liters of leri. Average yield of mineral content of potassium and their Duncan test results can be seen in **Figure 5**.

Giving yeast with a mass of 1 g per 5 liters of leri, was thought to cause the utilization of sugar in the medium to benot a limiting factor. So the mineral content of potassium in the medium remained high. It was also thought to acquire addition of mineral content of potassium from the microbe itself. According to Fardiaz (1989), the logarithmic growth phase of microbes were rapidly dividing and constant because of the availability of contained nutrients on the medium. On the contrary, at treatment of different fermentation period, the control non treatment had mineral content of potassium relatively higher and was probably as a result of the overhaul of potassium did not occur.

This study was the first step to prepare a liquid organic fertilizer which will be applied to plant to determine the effectiveness of leri-based organic fertilizers on growth and production of plant. The effect of nutritive minerals, vitamins, sugar alcohol and alcohols produced to plant growth were being determined.

4. Conclusion

Treatment of fermentation time of 6 days could produce relatively higher ethanol content, increasing the mineral content of phosphorus and sulfur, and also a good mineral content of nitrogen. Treatment of yeast concentration of 3 g per 5 liters of leri, produced better ethanol content and could increase the mineral content of nitrogen and phosphorus also has a good mineral content of potassium.

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