Identification Bacterial Genus from Floating Net Sediment in Tondano Lake in Biology Learning Of Biology Student in FMIPA UNIMA

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Abstract

The research was conducted to identify bacteria genus from sediment at fisheries floating net system in Tondano Lake. The method was use is descriptive method. Sediment sampel used to indentified was take from four location that is Remboken, Kakas, Eris and Tolour. Charateristic was to know are bacterial mophological in cultivated medium and biochemistry test. Determination the bacterial genus was use Bergey's manual of determinative Bacteriology 9th edition. The research result showed four bacterial genus found in floating net sediment that is Salmonella, Escherichia, and Enterbocater. It is stong recomendated to identify the species from four genus in the next research. Product of research used to biology learning. This kind of learning help the lecture to improve the learning sintax. Due to the student's respons, to improve the experimental pattern that force to reach the successful learning, because beside to increase high order thinking and also satisfaction of the student in order to create the enjoyment of learning.

Keywords: Identification, bacteri, genus, sediment, Tondano Lake, learning of biology .

1. Introduction

Lake Tondano has strategic role in Minahasa, and even in North Sulawesi. Besides as water reservoir for River Tondano that is exploited for PDAM Manado, PLTA Tanggari I and II, it is also exploited by the local communities for net system fish farming (floating net) in both lake and Tondano watershed (DAS). The other additional function is water reservoir for rice fields, duck farms, lake recreation and tourism and agricultural activities.

Net system fish aquaculture (floating net) in Lake Tondano approximately amounts to 3500 units. The need of fish feed is about 200-300 tonnes per month data Minahasa Department of Fisheries and Marine 2006. There is a growing perception of the fish farmers that feeding in large doses will lead to more rapidly fish growth so that the use of feed is more than the recorded data by the Minahasa Department of Fisheries and Marine. Fish feed as the given organic material increase the lake sedimentation rate and also causes the lake eutrification rate high (Kumurur, 2004). The abundance of nutrients lead to the abundant of aerobic and anaerobic bacterias both on the body of water and the sediment that formed at the lake bottom area of the net system fish farming (Sunarto, 2005).

About 19 bacterias are identified under the fish farming area that is also carried by the particular introduction of fish into the lake Tondano. The same thing was stated by Sumampouw (2007) that there are about 3 genus of bacteria in fish farming areas around the lake Tondano, i.e. Salmonella, Escherichia, Enterbocater and there are others genus which are not identified yet. So, it is necessary to conduct further study and continuous identification at certain time intervals, considering the existence of fish farming on the lake that change continuously.

Rapid bacteria growth especially in the fish farming area of Lake Tondano can lead to the development of various types of diseases that attack both human as well as fish and other organisms that may also exist in the lake. Identification of the types of exist bacteria is very important and the main thing to be implemented related to the existence of the Department of Biology, University of Manado (UNIMA) that surround the lake, where the result is the input to be known by the relevant agencies and have an impact on the subsequent management of the net system itself. Although there are harmful bacterias but there are also bacteria that can be used as decomposers of organic material, for example the organic matter in the sediment (Sunarto, 2005) which can be applied to the management of organic waste. Organic waste management is necessary and urgent, considering the rapid acceleration of lake silting. In 2008 the author had conducted depth measurements at several areas of the lakes associated with the mapping, and obtained results that the most depth in the area at the time was 24 m (in Tandengan area). Considering almost around the edge of the lake, the lake fishing activity that is fish aquaculture through nets is growing rapidly today.

The identification of bacteria genus which is at sediment of net system fish aquaculture in Lake Tondano helps to search the solution of today's Lake Tondano problems. And also in formal education side, relates to the location

of FMIPA UNIMA, it needs to conduct a study. Completing the identified work steps and its utilization can be used as material source especially in biology practicum of microbiology course. Cooperatively this acitivity can increase the scientific work and learning motivation, improving metacognitive skills and understanding of biological concepts, especially the solution of local natural resource use. It is same with Flavel, Brown and Livington in Sumampouw (2010) who suggest that metacognitive experiences involve metacognitive strategies or settings. This strategy is sequential process that is used to control cognitive activity and ensure that the cognitive objectives have been achieved. This process consists of planning, monitoring and evaluation of cognitive activity.

The research by Lawson et al. (2008) shows that learning effectivity can be increased if there is a task for the student to formulate scientific work involving cognitive activities. Due to this scientific reason, have developed models to be incorporated into an independent role in the study of biology S_1 with the development of scientific training models based on laboratory analysis of water samples. As a follow-up way of working in research, it requested a response in the form of evaluation that describes the response of concept understanding through the work done by the student. This activity is expected to help solving the lake problems, especially in microbiology, and utilize the learners to know more about the nature that is around.

Referring to the background described above, as for the study aims to identify the genus of bacteria in the sediment floating net system fish farming in the lake Tondano, responses, and responses of designed scientific workings by biology undergraduate FMIPA UNIMA students.

2. Research Method

The research is conducted in Department of Biology, Faculty of Mathematics and Natural Science UNIMA. There are 23 student of SBI class, Biology Education Department, Batch 2010 who contract the Microbiology class in academic year 2012, and are divided into 4 groups. The research locations are at FMIPA (Faculty of Mathematics and Natural Science) Microbiology Laboratory and North Sulawesi Health Departement Laboratory. Sediment samples are taken from several locations in the lake Tondano and specified in zig zag in order to obtain the area of floating net fish farming in Tandengan, Remboken, Kaweng and Paso. The used tools and materials are Olympus Binocular Microscope, glassware, autoclaves, laminar air flow, violet crystal, Lugol, safranin, NA media and distilled water.

The used method is descriptive. The research is conducted in some stages: 1) experiment stages designing in group; 2) the sampling using zig zag method according SNI; 3) bacteria isolation using dilution methods, i.e. 5 g of sediment sample is mixed with 45 ml of distilled water and it is blended. The found total colony is counted using a total plate count method; 4) dominant bacterial colonies in the further culture, morphology observation and biochemical tests (IMViC test, H_2S production and urease) then the determination using the referral key of determination Bergey's Manual of Determinative Bacteriology, 1994; 5) presentation of each design group, experiment results, and discussing results / obtained data between groups; 6) completing questionnaires as response / student response to each stage of the experiment.

3. Result and Discussion

3.1 Result

There are four stages of experimental design work in accordance with the existing number of groups referred to rubric which is presented in Table 1.

	Point					
Characteristic/Descriptor and point	Group 1	Group 2	Group 3	Group 4	Total (%)	Note
-Correct answer -Explain complete working stages -Elaborate clear systematics -point 4	1	-	-	-	25	
-Correct answer -Explain complete working stages -Elaborate less systematics -point 3		1	1	1	75	
-Less correct answer -Less complete working stages explanation -Unsystematic elaboration -point 2	-	-	-	-	-	
-Wrong aswer -Uncomplete working stages explanation - Unsystematic elaboration -point 1	-	-	-	-	-	

Table 1. The number of experimental design and the percentage of the group referred to the rubric.

The number of bacterial colonies on various sediment sampling locations of net system aquaculture in Lake Tondano is presented in Table 2.

Sample Source	The number of Bacterial Colonies on various level of dilution				Cell number /gr	
	10-1	10 ⁻²	10-3	10-4	10-5	
Tandengan	> 300	>49	<29	<20	<20	4200 cells/gr
Remboken	> 300	>300	<66	<56	<38	328.000 cells/gr
Kaweng	> 300	>300	<67	<57	<30	268.600 cells/gr
Paso	> 300	>82	<41	<41	<32	24.000 cells/gr

Table 2. The Number of Bacterial Colonies on Gelatin Nutrient Medium

The results of the morphological analysis and biochemical test of bacterial isolates are presented in Table 3.

Characteristics	Observed Colony			
	1	2	3	
1. Macroscopic				
ü Colony Form	Round	Round	Round	
ü Colony Color	Clear	Orange	Clear Yellow	
	transparant			
2. Macroscopic				
ü Cell form	Stick	Stick	Stick	
ü Gram	-	-	-	
ü Motalitas	+	+	+	
3. Biochemistry Test				
ü Indol	-	+	-	
ü H ₂ S	+	-	-	
ü Voges Proskaeur	-	-	+	
ü Metilen Red	+	+	-	
ü Urea hydrolysis	-	-	+	
ü Citric	+	-	+	
Conclusion	Salmonella	Escherichia	Enterobacter	

Table 3. Analysis of macroscopic, microscopic and Biochemistry

A positive Gram colony is found on the four sample areas with a yellowish color colony with uneven edges, round colony shape, stick form cells. Subsequent to the identification manually with reference to Bergey's Manual of Determinative Bacteriology, it is obtained that the colonies are the bacteria of the genus Bacillus (Holt et. al. 1994).

The characteristic of isolated result bacteria genus according to Holt et. al. 1994 as the following:

1. Genus Salmonela

The genus Salmonella is short stick bacteria. The cell size is 0.7 -1.5 μ m width and the length is 2-5 μ m, negative-gram, facultative aerobic, always motile with pentricus flagella. The optimum temperature is 30^oC. IMViC Indol and Voges-Proskauer test results are negative, methyl red and Simmon's test are positive citrate. Produce H₂S and does not hydrolyze urea. This bacterium is pathogenic bacteria.

2. Genus Escherichia

Genus Esherichia is in stem bacteria group. The cell size is $1.1 \,\mu$ m width and the length is $2.0-5.0 \,\mu$ m. Negativegram, facultative aerobic, motile with peritricus flagella or there are non-motile, the optimum temperature is 37^{0} C. IMViC test results, indole: positive, red positive methyl, negative Voges-Proskaeur and negative Simmon's citrate and not hydrolyze urea.

3. Genus Enterobacter

The genus Enterobacter is in bacteria groups whose stem cell size width 0.6 to 1 μ m and a length of 1.2 to 3 μ m. Negative-gram, motile with peritricus flagella, facultative aerobic. Optimum temperature is 37^oC. IMViC test results are negative indole, red methyl can be positive and negative, positive Voges-Proskaeur and do not hydrolyze urea.

In addition, data on student responses (%) on the workings of this study is presented in Table 4.

Table. 4 Student responses about this practicum						
No	Category	%	Note			
1	Really Like	24	Really Like			
2	Like	57	Like			
3	Less like	15	Less Like			
4	Dislike	4	Dislike			
	Total	100				

Table. 4 Student responses about this practicum

3.2 Discussion

Microbiology learning in this practicum shows the pattern of cooperative learning because working in groups and centering on the student activeness (student centered) in the sense that students can design their

practicum draft by themselves, after the lecturer suggested problems, goals and titles. Implementation of activities involves all members of the group, can work in groups while discussing practical activities, ranging from sampling of four groups, pre-sumtive tests, confirmation tests until the complete test phase in the laboratory. The steps of activities above are individual analysis activity that carried out in groups and this is very helpful to activate relevant knowledge to facilitate organizing and understanding of the subject matter so that the microbiology concepts can be arranged through the experiences. As noted by Corebima (2009), students in group work are known as the self-regulated learner, know when to perform assigned lab activities or when to consult with the supervisor (lecturer/assistant).

This pattern of cooperative learning is also done considering the principles of constructivism. The concepts of learning which is the characteristic of constructivist learning is believed to provide a significant opportunity for learners to construct their own understanding. As known, the keywords of constructivism learning philosophy is to construct. The observation accuracy and precision level of discovered concepts through this practicum activity determine the success in uncovering the observed phenomena and compile them into concepts. Contextual learning is essentially also meet the constructivism learning criteria, because one of the theoretical basis of contextual learning is the philosophy of constructivism. In addition to viewing learning as above to identify the genus of bacteria that exist in the lake Tondano, also can train critical thinking skills of learners. Livingston in Sumampouw (2010) stated that metacognitive holds a role to make successful learning and asserted that activities such as planning how to accomplish a specific task in learning, understanding and assessing monitoring task progress towards the completion of a task has a metacognitive nature. This exercise needs to be given to the learners, considering their existence later when are not in formal education anymore, they will be already familiar with the activity of metacognitive skills. It is very useful for learners in the era of globalization.

Genus Salmonella cannot produce enzim tripthophanase which is used for formation of indole from substrate triptopan when Kovac reagent is spilled on the medium; no color change (negative). Voges-Proskaeur is negative because Salmonella has no secretion of glucose into asetilmetilkarbonat so it does not react with alpha naptol and potassium hydroxide in the medium. However, Salmonella ferment glucose to acid when is spilled with red methyl so that the medium become red because it is in the acid solution (positive). Citrate test is positive because Salmonella utilize citrate as a carbon source in the growth. Salmonella does not produce urease so that it can not break down urea into ammonia and CO_2 (negative). Salmonella produce H_2S because it reduces the sulfur content in the medium so that the H_2S test result is positive.

The number of net system (floating net) fish aquaculture in coastal lakes Tondano cause an H_2S and ammonia increase in sediment and water bodies of net system fish aquaculture locations. The remains of fish feed and metabolism increase the content of H_2S and ammonia. These two compounds are highly toxic to fish and other organisms even in the lake which also has negative implications for coastal community lake using lake water for various purposes, most especially in fish farming activities.

These two compounds are very toxic for fish and other biota in lake that also give negative implication for the lake coastal communities who use lake water for various needs, especially in fish aquaculture activities.

Some species of bacteria are known to be pathogenic bacteria because it can cause illness to humans, e.g. diarrhea and even typhus. In the wild Salmonella can survive in water, soil and even groceries, feces, in humans up to two months (Entjang, Korua, 2003). Reported in 1997 that about 367 people in Remboken district who live in coastal lake suffered from diarrhea due to utilizing direct and indirect water lake. It seems that this problem should be solved, considering in current and future this lake is the livelihood source of people who living around the lake. With the detection of dangerous genus in the area of fish farming, it can be a concern of the government, society and so on to find an exact formulation, to find a solution that is acceptable to all concerned that the preservation of the lake is maintained.

The genus Escherichia produces enzim tripthophanase that can form indole from triptopan substrate. Similar with Salmonella, there is no glucose secretion which become into acetyl-methyl-carbonate so it does not react with alpha naptol and potassium hydroxide in the medium. It proves that Escherichia ferment sugars when it is droped with methyl red indicator so the medium becomes red in acidic conditions. Escherichia can also decipher citrate as a carbon source. Escherichia does not produce urease to catalyze urea into ammonia and CO_2 . Escherichia does not reduce the sulfur content so that it does not produce H_2S . Escherichia existence in the domestic environment is common for Escherichia can live in the intestines of humans and animals and is shipped on the feces. Communities around the lake are often known to take use the lake as a toilet.

Enterobacter also does not produce enzim tripthophanase which is used to form indole from triptopan substrate when Kovac,s reagen is spilled on the medium; there is no color change (negative). In contrast to the two previous genuses, the Proskaeur-Voges test is positive because Enterobacter secretea glucose into acetyl-methyl-carbonate that reacts with alpha naptol and potassium hydroxide indicated by the color changing of the medium

which is overgrown with bacteria to red. Methyl red test show negative result because it cannot ferment glucose. While the citrate test is positive because Enterobacter utilize citrate as a carbon source in the growth. Enterobacter can catalyze urea into ammonia in the presence of urease enzyme which is generated by it. Produce H_2S because reduce the sulfur content in the medium.

Bacteria in Enterobacter genus are cosmopolitan because it has high tolerance to various environmental conditions. Enterobacter is often found in various waste water, soil, human and animal feces. This genus is a bacteria genus that live on the ground, but also found in fresh waters (Hendrati, 2003). This genus has a high resistance to the environment and can survive for long, some species cause infections in humans and animals (Pelczar and Chan, 1999). In addition to identifying bacteria that exist in the lake Tondano floating net system sediment, it also obtains feedback / response about the student's learning.

Most of the student responses on practicum learning model like this is really like and dislikes. Taking into account the response and motivation of learners on learning models such as this, means more empowering learners to the change towards constructivism learning process with contextual basis. Ask students what can be done about this kind of feedback is very helpful for learning tutors / lecturers in improving learning syntax. Reffering to the student's response then fix the experimental pattern for this topic seems especially pressing toward successful learning, because in addition to increase the student understanding of the concept, it is also a satisfaction in creating a fun learning. In this era of science development, to be able to contribute and be able to follow the developments in the field of microbiology, requires the fun scientific work skills.

5. Conclusion

Learning through design their own practicum in group shows the pattern of cooperative learning that is centered on the student (student centered) and the work of students in the group known as the self-regulated learner.

Based on the bacterial genus identification result in the area of net system (floating net) fish aquaculture sediment in the lake Tondano; it may be concluded that there are three bacteria genuses, i.e. the genus Salmonella, Escherichia, Enterobacter. Students who enjoy learning by designing their own experiments amounts to 81% and 19% of them less like it.

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