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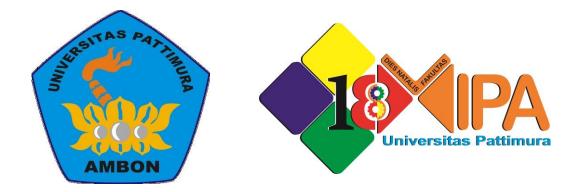
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## PROCEEDINGS

## The 2<sup>nd</sup> International Seminar of Basic Science

"Natural Science for Exploration The Sea-Island Resources"

Poka-Ambon, 31<sup>st</sup> May 2016

Mathematic and Natural Science Faculty Universitas Pattimura Ambon 2016

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## Welcoming Address By The Organizing Committee

Today, We have to thank the The Almighty Allah SWT for the implementation of this international seminar. This is the second seminar about Basic Science in The Faculty of MIPA Pattimura University. The seminar under the title "Natural Sciences for Exploration the Sea-Island Resources" will be carried out on May 31<sup>st</sup> 2016 at Rectorate Building, Pattimura University. There are 200 participants from lecturers, research institute, students, and also there are 34 papers will be presented.

My special thanks refer to the rector of Pattimura University and the Dean of MIPA Faculty, Prof. Dr. Pieter Kakissina, S.Pd., M.Si. I also would like to express my deepest gratitude to Prof. Amanda Reichelt-Brushett, M.Sc., Ph.D. ; Kazuhiko Ishikawa, Ph.D. ; Nicolas Hubert, Ph.D. ; Prof. Dr. Kirbani Sri Brotopuspito ; Prof. Dr. Marjono, M.Phil. ; Gino V. Limon, M.Sc., Ph.D. as the keynote speakers.

The last, We hope this international seminar usefull for all of us, especially Mollucas People and very sorry if any mistake. Thank you very much.

#### Dr. La Eddy, M.Si.

Chairman of Organizing Committee

## Opening Remarks By Dean of Mathematic and Natural Sciences Faculty

I express my deepest gratitude to The Almighty God for every single blessing He provides us especially in the process of holding the seminar until publishing the proceeding of International Seminar in celebrating the 18<sup>th</sup> anniversary of MIPA Faculty, Pattimura University. The theme of the anniversary is under the title "Natural Sciences for Exploration the Sea-Island Resources". The reason of choosing this theme is that Maluku is one of five areas in Techno Park Marine in Indonesia. Furthermore, it is expected that this development can be means where the process of innovation, it is the conversion of science and technology into economic value can be worthwhile for public welfare especially coastal communities.

Having the second big variety of biological resources in the world, Indonesia is rich of its marine flora and fauna. These potential resources can be treated as high value products that demand by international market. Basic science of MIPA plays important role in developing the management of sustainable marine biological resources.

The scientific articles in this proceeding are the results of research and they are analyzed scientifically. It is expected that this proceeding can be valuable information in terms of developing science and technology for public welfare, especially people in Maluku.

My special thanks refer to all researchers and reviewers for your brilliant ideas in completing and publishing this proceeding. I also would like to express my gratefulness to the dies committee-anniversary of MIPA Faculty for your creativity and hard working in finishing this proceeding, God Bless you all.

#### Prof. Dr. Pieter Kakisina, S.Pd., M.Si.

Dean of Mathematic and Natural Sciences Faculty

## ACKNOWLEDGMENT

The following personal and organization are greatfully acknowledgment for supporting "The 2<sup>nd</sup> International Seminar of Basic Science 2016"

Hotel Mutiara Ambon

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#### HISTOLOGICAL OF HAEMOCYTE INFILTRATION CHANGES DURING PEARL SAC FORMATION IN *Pinctada maxima* HOST OYSTERS REARED AT DIFFERENT DEPTHS

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#### ABSTRACT

This study was designed to determine the effect of different depths of rearing on the haemocytes infiltration in *Pinctada maxima* oyster. *Saibo* used was aged 28 months *Pinctada maxima* oyster, nucleus position in the ventral gonad and using female oysters. The first factor was the depth of rearing consisted of 4 levels i.e., 3, 6, 9, and 12 meters. The second factor was time after implantation with 4 levels i.e., 1, 2, 3, and 4 weeks. The results of histological observations showed that the haemocytes and inflammatory cells infiltrations were similar in the *Pinctada maxima* host oysters of the depth of rearing 3, 6, 9 and 12 meters. It was concluded that haemocyte infiltration in oysters reared at the depth of 3, 6, 9 and 12 meters no differences.

Keywords: Depth of rearing, Pinctada maxima, Pearls sac formation

#### INTRODUCTION

Pearl oyster survival is influenced by exogenous factors, such as temperature, a period of months, depth, abundance, mechanical factors, the availability of food, and light intensity, and endogenous factors such as genetic and hormonal (Wada 1984).

Depth is associated with the availability of food, salinity, and temperature. Temperature is one of the factors that may affect survival, including growth and reproduction (Helmuth *et al.* 2006; Menge *et al.* 2008). Salinity also affects the growth of pearl oysters. Stress hiperosmotic make changes to the system of osmoregulation and osmotic signal that ultimately affect gene expression (Lockwood & Somero 2011; Zhao *et al.* 2012; Meng *et al.* 2013). Stress salinity affects calcium signal transduction pathways (Zhao *et al.* 2012; Meng *et al.* 2013).

These data and information are essential in understanding the biological basis of the effect of the different depths of rearing on the haemocytes infiltration in *Pinctada maxima* oysters.

#### MATERIALS AND METHODS

#### **Experimental Materials**

This study was conducted from July 2012 to November 2012 at the commercial pearl farm of CV. Aru Duta Indah in the Garaga, Obi Island (01°25'S, 127°20E) North Moluccas Province, Indonesia. Host oysters used in this experiment were *Pinctada maxima* oysters that were cultured by the commercial pearl farm. The first factor was the depth of rearing consisted of 4 levels i.e., 3, 6, 9, and 12 meters. The second factor was time after implantation with 4 levels i.e., 1, 2, 3, and 4 weeks. Forty eight (48) (4 x 4 with 3 replications) oysters succeeded in implantation were used for measurement of histological haemocyte infiltration during pearl sac development.

*Pinctada maxima* oysters used in the experiment as hosts were selected by criteria of normal morphology (without shell malformation), the same level of gonad maturity (gonadal development phase), with similar dorso ventral margin (DVM) of 12 cm and anterior-posterior margin (APM) of 11 cm, and body weight ranged of 180-210 gram.

#### **Parameters Measured**

The histological preparation of the haemocyte infiltration during pearl sac developing pearl sac used the haematoxcillin eosin staining (HE) technique. Data analysis was done descriptively.

#### **RESULTS AND DISCUSSION**

#### Results

The effect of 3 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters presented in Fig.1. The inflammatory cells and haemocytes infiltrations were high 1 week after implantation. Two weeks after implantation, the number of inflammatory cells and haemocytes began to decline. Three and four week after implantation, no haemocytes and inflammatory cells was found.

The effect of 6 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters presented in Fig.2. One week after implantation, the inflammatory cells and haemocyte were high. Two weeks after implantation, the number of inflammatory cells and haemocyte began to decline. Three weeks after implantation, the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation, there was no haemocyte and inflammatory cell was found.

The effect of 6 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters presented in Fig.2. One week after implantation, the inflammatory cells and haemocyte were high. Two weeks after implantation, the number of inflammatory cells and haemocyte began to decline. Three weeks after implantation, the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation, there was no haemocyte and inflammatory cell was found.

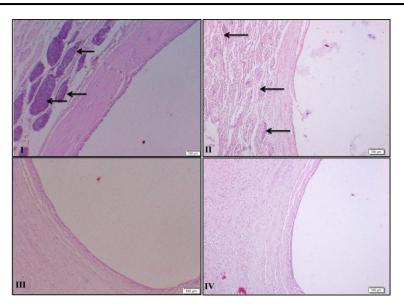
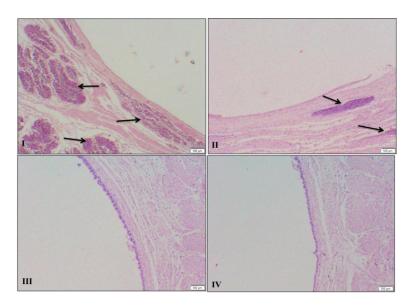


Figure 1. The effect of 3 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters. Arrows indicated haemocytes. One week after implantation (I), inflammatory cells and haemocytes were high. Two weeks after implantation (II), the number of inflammatory cells and haemocytes began to decline. Three and four weeks after implantation, there was no haemocyte. H&E. x100.



**Figure 2.** The effect of 6 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters. Arrows indicated haemocytes. One week after implantation (I), the inflammatory cells and haemocyte were high. Two weeks after implantation (II), the number of inflammatory cells and haemocyte began to decline. Three weeks after implantation (III), the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation (IV), there was no haemocyte and inflammatory cell was found. H&E. x100.

The effect of 9 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters presented in Fig.3. One week after implantation, haemocyte were high. Two weeks after implantation, haemocyte began to decline. Three weeks after implantation, the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation, there was no haemocyte and inflammatory cell was found.

The effect of 12 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters presented in Fig.4. One week after implantation, the inflammatory cells and haemocyte were very high. Two weeks after implantation, the number of inflammatory cells and haemocyte began to decline. Three weeks after implantation, the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation, there was no haemocyte and inflammatory cell was found.

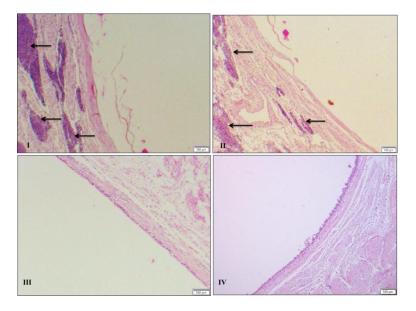


Figure 3. The effect of 9 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters. Arrows indicated haemocytes. One week after implantation (I), haemocyte were high. Two weeks after implantation (II), the haemocyte began to decline. Three weeks after implantation (III), the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation (IV), there was no haemocyte and inflammatory cell was found. H&E. x100.

### PROCEEDINGS

The 2<sup>nd</sup> International Seminar of Basic Science May, 31<sup>st</sup> 2016

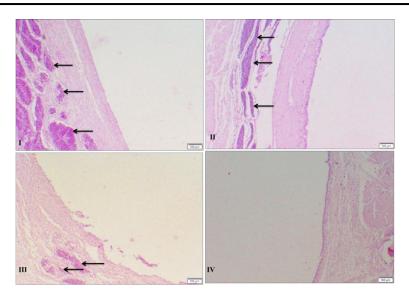


Figure 4. The effect of 6 meters depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters. Arrows indicated haemocytes. One week after implantation (I), the inflammatory cells and haemocyte were very high. Two weeks after implantation (II), the number of inflammatory cells and haemocyte began to decline. Three weeks after implantation (III), the number of inflammatory cells and haemocytes were very low and the injury began to recover. Four weeks after implantation (IV), there was no haemocyte and inflammatory cell was found. H&E. x100.

#### Discussion

This results that effect of diferences depths of rearing on the haemocytes infiltration change during pearl sac formation in *Pinctada maxima* oysters as the same. There was no haemocyte and inflammatory cell was found 4 weeks after implantations. This result indicated diferences depths of rearing in *Pinctada maxima* oysters no affect. Operation process during nucleus implantation caused the oyster stress. Stress increases the total haemocytes on the *Crassostrea gigas* oyster. Stress affects several hormones activities such as CRH (corticotrophin releasing hormone), ACTH (adrenocorticotrophic hormone), cytokines, noradrenaline, adrenaline, dopamine, and cortisol (Lacoste *et al.* 2002). Stress activates the endocrine system such as corticotrophin releasing hormone (ACTH). The presence of ACTH stimulates the release of adrenocorticotrophic hormone (ACTH). The presence of ACTH stimulates the release of biogenic amino acids, which eventually lead to secondary effects on oysters (Hooper *et al.* 2007).

Exogenous factors, such as depth and mechanical factors no effect in infiltration haemocytes, (Hooper *et al.* 2007) that was associated with the increased glucose concentrations. Increased stress during early implantation increased haemocyte infiltration and haemolymph glucose concentration. When the implantation injury was cured, haemocytes was low and haemolymph glucose concentration reached the lowest levels. The decreased haemolymph glucose concentration with the advance of pearl sac growth after implantation could indicate the possibility of increased glucose uptake without increased in glucose mobilization or uptake to the haemolymph. Glucose are required for energy source for basal metabolism and for supporting synthetic activities as well as for synthesis of

material build up from glucose, such as conchiolin. Conchiolin is organic in nature and consists of mucopolysaccarides (Chellam *et al.* 1991).

#### CONCLUSION

Haemocyte infiltration during pearls sac formation in *Pinctada maxima* oysters reared at the depth of 3, 6, 9 and 12 meters no differences.

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