

BOOKS OF THE ABSTRACTS

NSYSU-KU Seminar on Agriculture and Fishery Sciences

Kaohsiung, Taiwan

October 24, 2013



Kagoshima-Maru : Maiden Voyage to the Port of Kaohsiung

National Sun Yat-sen University

Kagoshima University



日本鹿兒島大學研究船首航高雄

NSYSU-KU 農業及漁業科學研討會論文摘要集

Abstracts of Contributions Presented at the NSYSU-KU Seminar on
Agriculture and Fishery Sciences, Kaohsiung, Taiwan
October 24, 2013

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國立中山大學海洋科學院

AGENDA

October 24, 2013 (Thursday)

Lecture Hall, College of Marine Sciences

National Sun Yat-sen University

13:00~13:15	REGISTRATION
13:20~13:30	OPENING OF THE SEMINAR (Prof. Hin-Kiu Mok) Prof. Hounng-Yung Chen (College of Marine Sciences, National Sun Yat-sen University) Prof. Tatsuro Matsuoka (Faculty of Fisheries, Kagoshima University)

SESSION 1	Chairperson : Dr. Kazuhiko Anraku	Page Number
13:30~13:42	Utilizing target strength measurements to estimate fish size distribution in Kagoshima Bay <i>Berry Muller</i> Faculty of Fisheries, KU	1
13:42~13:54	Basic study on hygiene management using electrolyzed water in a shochu factory <i>Uchida Keiichi</i> United Graduate school of Agricultural Sciences, KU	3
13:54~14:06	Sinking of the oceanic free-living heterocystous filamentous cyanobacteria <i>Sing-how Tuo</i> Department of Marine Biotechnology and Resources, NSYSU	5
14:06~14:18	Food sources for benthos in shallow hydrothermal vent and non-vent ecosystems <i>Jing-Ying Wu</i> Department of Oceanography, NSYSU	7
14:18~14:30	A study on agroforestry in private forest and its contribution to farm economy in Indonesia -A case study of sukaresmi village in Cianjur, West Java <i>Chiharu Maeda</i> United Graduate school of Agricultural Sciences, KU	9

14:30~14:42	Study on coastal groundwater <i>Takashi Kamo</i> United Graduate school of Agricultural Sciences, KU	11
14:42~14:54	Effects of dietary fatty acids on cell differentiation and lipid accumulation of preadipocytes in cobia (<i>Rachycentron canadum</i> Linnaeus) <i>Yu-Chun Cheng</i> Department of Oceanography, NSYSU	13
14:54~15:06	Catch composition of tropical tuna associated with floating objects and free schools by purse seine in the Indian Ocean <i>Watcharapong Chumchuen</i> United Graduate school of Agricultural Sciences, KU	15
15:06~15:18	Biological control of pathogenic root rot fungus <i>Hongyi Wei</i> United Graduate school of Agricultural Sciences, KU	17

15:18~16:00 COFFEE BRAKE AND POSTER SESSION

SESSION 2 Chairperson : **Dr. Kazuyoshi Komeyama**

16:00~16:12	Age and size effects on total and organic mercury concentrations in the white muscle of North Pacific albacore <i>Kuo-Shu Chen</i> Department of Oceanography, NSYSU	19
16:12~16:24	Warming induced inconsistencies in ocean acidification rates based on pH ₂₅ and pH _{insitu} <i>Hon-Kit Lui</i> Department of Oceanography, NSYSU	21
16:24~16:36	Reproductive seasonality of <i>C. annulus</i> (Mollusca: Gastropoda) in Hanazesaki, Kagoshima Prefecture, Japan <i>Shiela Villamor</i> United Graduate school of Agricultural Sciences, KU	22
16:36~16:48	Sponge diversity in the sea grass bed of Dongsha Island, Taiwan <i>Yalan Chou</i> Institute of Oceanography, NSYSU	24

16:48~17:00	Introduction of miso in Japan and manufacturing method <i>Toshihiro Kojo</i> United Graduate school of Agricultural Sciences, KU	25
17:00~17:12	Response of fishers toward application of fishery management and development initiatives. Sub-title: A comparative study on responses of fishers of Tuvalu, Japan and Malaysia <i>Nikolasi Apinelu</i> Faculty of Fisheries, KU	27
17:12~17:24	Development of new methodology to calculate gillnet selectivity curve and estimation of population length distribution of pink salmon (<i>Oncorhynchus gorbuscha</i>) <i>Aynealem Goitom Berhe</i> Faculty of Fisheries, KU	29
17:24~17:36	Climate effects on temporal variation in abundance and distribution of the demersal fish assemblage in the Tsushima Warm Current region of the Japan Sea <i>Chen-Yi Tu</i> Institute of Oceanography, NTU	31
17:36~17:48	Population structure of Limpid-wing flyingfish (<i>Cheilopogon unicolor</i>) in the north-western Pacific inferred from mitochondrial CO I genes <i>Chang-En Chou</i> Department of Biological Science, NSYSU	33
17:48~17:55	CLOSING OF THE SEMINAR <i>Prof. Hin-Kiu Mok</i> (College of Marine Sciences, National Sun Yat-sen University)	

Poster Session

Lecture Hall Hallway, College of Marine Sciences

National Sun Yat-sen University

NUMBER	AUTHOR	TITTLE	PAGE NO.
P01	Yuya Akamatsu	Development of the LED fishing light based on the visual sensitivity of fish	35
P02	Shintaro Manago	Dynamics of subtropical zooplankton is steady-state? As an evidence of seasonal changes in population structure and depth distribution of two subtropical copepods	37
P03	Sachi Miyake	Nucleic acid ratios can be a proxy of egg production rate for marine copepods?	39
P04	Rie Nakamura	Analysis of trophic dynamics in zooplankton community using stable isotope ratio	41
P05	Satoshi Kuwabara	Effect of towing time for bottom trawl on freshness of the shrimp	43
P06	Yusuke Yamashita	Evaluation on the freshness of catches between catching and loading for stick held dip net fishery	45
P07	Kazunari Tanaka	Analysis of turning behavior of reared Pacific Bluefin tuna in an aquaculture net cage	47
P08	Bo-Wei Su	A comparative study on the development of giant clams <i>Tridacna noae</i> and <i>Tridacna maxima</i> (Bivalvia: Tridacnidae) in Taiwan	49
P09	I-Chun Lin	Reproduction and recruitment of land crabs (Decapoda: Brachyura) in Hsiangchiaowan, Hengchun Peninsula, Taiwan	51
P10	Hsu-Sen Chen	Reproductive and migrations of the southern velvet shrimp <i>Metapenaeopsis palmensis</i> in the waters off southwestern Taiwan	52

P11	Yousuke Taoka	Influences of light irradiation on production of fatty acids and carotenoids in thraustochytrids, genus <i>Aurantiochytrium</i>	53
P12	J.T. Liu	From the highest to the deepest: A river-sea dispersal system on the edge of the South China Sea	
P13	R.J. Yang	EOF analysis of the continuous core data during the late Quaternary from the Zhuoshui River Delta	
P14	J. Li	River plume related variability of suspended sediment characteristics off the mouth of Zhuoshui river on the west coast of Taiwan	
P15	R.T. Hsu	The observation of hyperpycnal turbidity current in the Gaoping Submarine Canyon in the wake of the Typhoon	
P16	Pa-Ching Song	Cloning and characterization of ApRab4, a recycling Rab protein of <i>Aiptasia pulchella</i> , and its implication in the symbiosome biogenesis	
P17	Yu-Chun Cheng	Effects of 18-carbon fatty acids on adipogenesis and PPARs expression in cobia preadipocytes	
P18	Jing-Ying Wu	The spatio-temporal variations in food source of the crab <i>Xenograpsus testudinatus</i> from shallow-water hydrothermal vents, Kueishan island	
P19	Chiu-Chin Lu	Antimicrobial peptides extract from the marine benthic diatoms	
P20	Jian-Xiang Liao	How many replicates are required to represent the local meiofauna diversity in tropical seagrass beds?	
P21	Ming-Chang, Hong	A Vacuolar (H ⁺) ATPase-like involved in phagosomal acidifying is exclude from symbiosomes between Aiptasia-Symbiodinium endosymbiosis	
P22	Yi-Reng Lin	Comparative proteomics analysis of degenerative eye lenses of nocturnal rice eel and catfish as compared to diurnal zebrafish	

P23	Che-Wei Hu	Characterization of a salt-induced reddish microalga: isolated microalga F50 and its potential on biofuel and carotenoids production
P24	Jen-chun Hsiue	Compare sound characteristics and sonic structures in <i>Terapontidae</i> and <i>Pempheridae</i>
P25	Titisari Dian Pertiwi	Comparative study on the sonic muscles of <i>Glaucosomatid</i> , <i>Pempherid</i> , <i>Terapontid</i> , and <i>Ophidiid</i> : a proteomic approach

Editor

Yu-Chun Cheng

**Department of Oceanography,
National Sun Yat-sen University**

Utilizing target strength measurements to estimate fish size distribution in Kagoshima Bay

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1. Purpose

Fish detection by acoustics means were first trialed during the 1930s and have since become an important part of commercial fisheries. Furthermore, underwater acoustics is also becoming an increasingly useful tool in the assessment and management of fisheries resources (Simmonds & MacLennan, 2005).

The target strength (TS) reference value is an important requirement for biomass estimation using acoustics as it provides a sample representation of the population. In this research, the individual TS data were collected and will be extrapolated through statistical methods to estimate fish size distribution by location, seasonality and depth stratification within Kagoshima Bay.

One of the main issues facing fisheries today is the adverse impacts of tuna purse seine fishing using fish aggregating devices (FADs). There is growing concern by the Western and Central Pacific Fisheries Commission (WCPFC) about the high fishing mortality rates on small bigeye and yellowfin tunas resulting from this type of fishing activity (WCPFC, 2012). Although the bigeye tuna stock is targeted mostly by the longline fishery, purse seine catches of the juvenile component of the stock has increased steadily with the introduction of FADs in the late 1990s.

The purpose of the research is to gain a better understanding of the fish size distributions using acoustic methods. The intention is that this type of research can be a basis for further studies conducted during actual fishing operations and provide advice to fishers on possible mitigation measures that may address catches of juvenile bigeye tuna and other bycatch species associated with the tuna fishery.

2. Materials and Methods

During 2012-2013, acoustic data were collected at 6 different time intervals onboard the R/V Nansei Maru in Kagoshima Bay, a semi-enclosed body of water in southern Japan. The research vessel is equipped with a split-beam quantitative echosounder (KFC-3000, KAIJO) operating at a frequency of 38 kHz. Ongoing post processing continues to be undertaken via the Echoview 5 software and SPSS cluster analysis to examine any patterns in mean fish sizes both in different sections of the bay and by depth strata.

3. Discussion

As the data analysis is still in initial stages, the results are yet to be available for evaluation.

However, the application of underwater acoustics in fisheries and continued improvements to such instruments can potentially allow fishers to extract such information as fish size, species and abundance prior to fishing operations. With such knowledge, fishers can have the ability to avoid setting on fish schools with high levels of non-target species and under-sized fish.

Shaefer and Fuller (2007) suggest that underwater acoustics can play a role in a purse seine vessel captain's ability to reliably estimate the species composition, size frequency, and tonnage of tunas associated with floating objects before setting their nets. Such studies can lead to better refinement of acoustic methods in the future and provide more precise estimates to advise overall fisheries management.

4. References

- 1) WCPFC. 2012. Summary report of the ninth regular session of the Commission for the Conservation and Management of Highly Migratory Fish Stocks in the Western and Central Pacific Ocean. 2-6 December 2012, Manila, Philippines.
- 2) Simmonds, J. & MacLennan, D. 2005. Fisheries acoustics: theory and practice (2nd ed). Blackwell Science, London.
- 3) Shaefer, K. & Fuller, D. 2007. Acoustic imaging, visual observations, and other information used for classification of tuna aggregations associated with floating objects in the Pacific Ocean. Interamerican Tropical Tuna Commission, La Jolla, California, USA. Western and Central Pacific Fisheries Commission Scientific Committee Third Regular Session, 13-24 August 2007, Honolulu, USA. WCPFC-SC3-FTSWG/WP-2.

Basic study on hygiene management using electrolyzed water in a “Shochu” factory

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1. Abstract

The safety of food are requirements of consumer. Therefore, many sterilizers are used in food processing to keep cleanness of processing machines. However, using a lot of detergent causes environmental pollution. The food processing companies need new sterilizers which do not cause environmental pollution and have sufficient washing and bactericidal effect. In this study, it was found that using electrolyzed water led to little environmental pollution, additionally the effectiveness of cleaning and disinfection in production process of sweet potato “Shochu” (Fig.1).



Fig. 1 manufacturing process of sweet potato

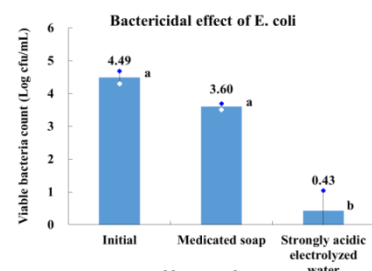


Fig. 2 Bactericidal effect of medicated soap and strongly acidic electrolyzed water.

2. Use of acid electrolysis water -Verification of the bactericidal effect in fingers- Background and purpose

Since *E. coli* in human hands contaminated the process line, quality degradation such as flavor degradation of sweet potato “Shochu” occur red. However, use of the agents for hand-washing of workers or the procedure has not been fixed. In this experiment, the bactericidal effect of strongly acidic electrolyzed water and conventional methods of hand-washing were compared to verify the possibility of using electrolysis as an alternative treatment.

Results

The bactericidal effect of the strongly acidic electrolyzed water on *E. coli* was stronger than medicated soap ($P<0.01$) (Fig.2). It is possible to use acid electrolyzed water as an alternative to medicated soap.

3. Use of alkaline electrolyzed water -Verification of washing effect in stainless steel tank-Background and purpose

Potato fat (Jalapin) adheres to the stainless steel tanks in the manufacturing process of sweet potato “Shochu”. Potato fat remaining in the stainless steel tank affects the quality of the “Shochu”. A neutral detergent and alcohol are used for washing in the factory. In this experiment, the washing effects of alkaline electrolyzed water and conventional methods (Ethanol 70% + Keiporu 2%) are compared to verify the possibility of alternative.

Experimental methods

In the stainless steel tank contaminated with potato fat, the degree of contamination was different in the upper and the lower area. For this reason, the tank was divided into four parts (Fig.3) and washed with different cleansers. The cleaning level was quantified with the ATP method.

Results

Washing effect of the conventional method and alkaline electrolyzed water was similar in the upper area. Washing effect was higher in the alkaline electrolyzed water than in the conventional methods in the lower area ($p > 0.05$). It is possible to use alkaline electrolyzed water as an alternative to the conventional method.

4. Conclusion

It is possible to use alkaline electrolyzed water for sterilizing hands of the employees and for washing the equipment contaminated with the potato fat as an alternative to the conventional methods. Alkaline electrolyzed water is safe for the environment and people, thus we expect that the use of alkaline electrolyzed water will increase in various fields in the future.

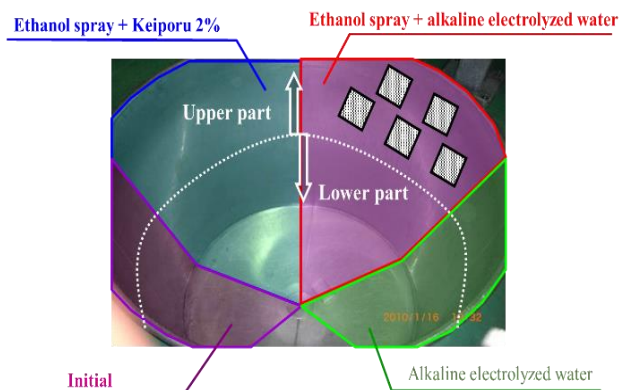


Fig. 3 Verification of washing effect of alkaline electrolyzed water and conventional methods on potato fat.

Sinking of the oceanic free-living heterocystous filamentous cyanobacteria

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1. Purpose

Richelia intracellularis and *Calothrix rhizosoleniae* are the most important N₂-fixing heterocyst-bearing filamentous cyanobacteria in nitrogen-limited tropical oceans. They frequently symbiosis with host diatoms *Rhizosolenia*, *Hemiaulus*, *Chaetoceros*, and *Bacteriastrium*. Free-living *Richelia* and *Calothrix* (called as free-living cyanobionts, FC) are rare in nature. *Richelia* have no gas vesicles (Janson et al. 1995), which implies that they might sink faster than their host diatoms possessing mechanisms to regulate their buoyancy. Theoretically, FC with longer filaments sink faster than those with shorter filaments according to Stokes' law. If this is the case, most FC in the upper euphotic oceans would have short filaments. To elucidate this, we measured in the laboratory using an FC monoculture the sinking rates of FC with different filament lengths; and linked the results to the distribution of FC filament lengths in the nature population.

2. Materials and methods

The distribution of FC filament lengths in the natural population were measured in eight cruises in the cold and warm seasons in the northern South China Sea (SCS) and the Kuroshio. The filament lengths were classified as short (SFC, 2-5 cells), medium (MFC, 6-11 cells), and long (LFC, >11 cells). The sinking rates of FC for the three filament groups were determined by the Utermöhl method. The relationship between the sinking rate and the filament length was established. The obtained FC sinking rates were also used to compare with the literature-reported sinking rates of the potential host diatoms *Rhizosolenia*, *Bacteriastrium*, and *Chaetoceros*.

3. Results

Over 95% of natural population belonged to either SFC or MFC (Fig. 1), with a mean of 6 cells per filament. The occurrence of LFC was rare (2-3%) in the warm season in the SCS and both warm and cold seasons in the Kuroshio. The FC possessed relatively long filaments (LFC 19%; 9 cells per filament) in the cold season in the SCS. In the laboratory study, the sinking rate of FC was positively correlated with the filament length (Fig. 2). LFC sank significantly ($p < 0.05$) faster

than MFC and SFC. LFC also sank faster ($p < 0.05$) than diatoms *Rhizosolenia*, *Bacteriastrum*, and *Chaetoceros*, which are the potential hosts of FC.

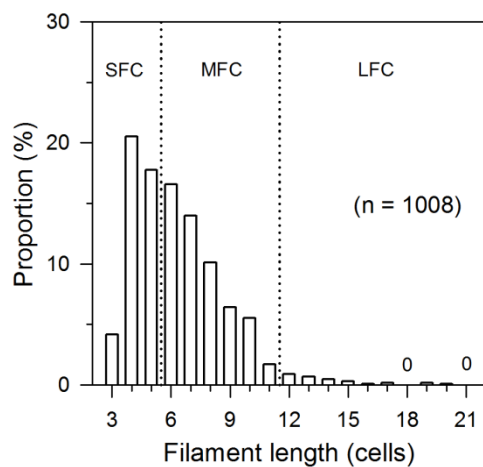


Fig. 1. Proportion of the free-living cyanobionts in various filament lengths (SFC 2-5 cells, MFC 6-11 cells, and LFC, >11 cells). n, number of filaments measured.

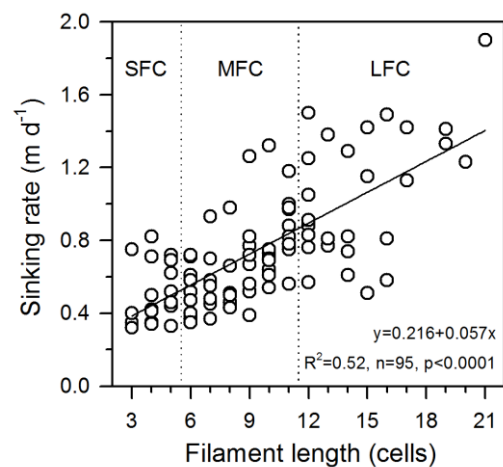


Fig. 2. Relationship between filament length and sinking rate of the free-living cyanobionts. n, number of experiments.

4. Discussion and conclusion

- (1) Very few LFC occurred in nature.
- (2) FC with a longer filament sank faster than that with a shorter filament.
- (3) LFC sank faster than the potential host diatoms, but not MFC or SFC.
- (4) On average in the cold season LFC occurred frequently in the SCS. The phenomenon might be related to the cyclonic eddy formed in the basin. The upwelling from the eddies could prevent the sinking of the FC.

Conclusions: the results suggested that the rareness of FC in nature could be the cumulative results of the greater sinking rates of LFC than SFC or MFC in the upper euphotic zone, leaving only FC with filaments of few cells.

5. References

- 1) Janson S, Rai AN, Bergman B (1995) Intracellular cyanobiont *Richelia intracellularis*: ultrastructure and immuno-localisation of phycoerythrin, nitrogenase, Rubisco and glutamine synthetase. Mar Biol 124:1-8

Food sources for benthos in shallow hydrothermal vent and non-vent ecosystems

Jing-Ying Wu*, Siou-Yan Lin, Ya-Jou Chen, Shao-Hung Peng, Jia-Jang Hung, Chen-Tung A.Chen, Li-Lian Liu

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1. Purpose

It has been reported that the primary food source is dead zooplanktons killed by sulphurous plumes in shallow vent ecosystems off Kueishan (KS) Island, Taiwan. In this particular vent system, the recorded macrofaunal species are a hexacoral *Tubastraea aurea*, an unidentified sea anemone, a snail *Nassarius* sp., an unidentified chiton, one serpulid polychaete, and the vent crab *Xenograpsus testudinatus* which is the predominant one. In the present study, food source signals of macrofauna were investigated by stable isotope approach and one dimension protein expression profiles.

2. Materials and methods

The sediment organic matters (SOM), zooplankton and benthic macrofauna were collected in vent and non-vent areas during 2009-2010. The samples for $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ determination were prepared by lyophilizing, grinding and weighted in tared tin cups. Stable isotope measurements were performed at the Stable Isotope Facility at University of California, Davis. The relative contributions of different food sources were analyzed by the Bayesian stable-isotope mixing model (SIAR).

In situ collected crabs and live specimens were brought to laboratory for the analyses of protein expression profiles through one dimensional sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE).

3. Results

The values of $\delta^{15}\text{N}$ (‰) and $\delta^{13}\text{C}$ (‰) of macroalgae and sediment organic matters (SOM) from vent areas were lower than that of tail-side. The $\delta^{15}\text{N}$ (‰) of the crab *X. testudinatus* from yellow vent was lower than those from white vent, while $\delta^{13}\text{C}$ (‰) was on the contrary. In contrast, signals in the copepod and fish were no difference among sites. Filter feeders, scavengers and carnivores were the major feeding types of vent benthos. Main food items for Calyptraeidae snail and chiton were SOM and zooplankton debris. The crab *X. testudinatus* from white vent were primarily fed on mollusks and it was periphyton for yellow vent crabs based on the results of Stable Isotope Analysis in R (SIAR). Furthermore, the protein expression profiles

showed that there was a little difference between crabs from different vents. After culture in laboratory for 12 hours, crab protein expression profiles were different from their original ones, indicating crab response to new environment was relatively fast.

4. Discussion

- 1). Inorganic carbon source for macroalgae and benthic microbia is probably from hydrothermal fluid based on the $\delta^{13}\text{C}$ signals.
- 2). SOM in vent areas might contain organisms which are responsible for chemosynthetic assimilation of ammonium.
- 3). Marine 'snow' is the food source for chiton, polychaete, sea anemone and the coral *Tubastrea aurea*. Their stable isotope signals are closed to zooplanktons with a 1 ‰ shift of $\delta^{13}\text{C}$ and 3.4 ‰ shift of $\delta^{15}\text{N}$.
- 4). Crabs with different protein expression profiles indicate that they can adapt toxic and non-toxic environment quickly.

5. References

- 1) Jeng, M.-S., Ng, N. K. & Ng, P. K. L. Hydrothermal vent crabs feast on sea snow. *Nature* **432**, 969 (2004).
- 2) Inger, R., Jackson, A., Parnell, A. & Bearhop, S. (2008) SIAR V4 (Stable Isotope Analysis in R) An Ecologist's Guide.
http://www.tcd.ie/Zoology/research/research/theoretical/siar/SIAR_For_Ecologists.pdf
- 3) Chang, C. M. Sulfide tolerance and detoxification of the vent crab, *Xenograpsus testudinatus*. Master Thesis, Institute of Oceanography, National Taiwan University. pp. 49

A study on agroforestry in private forest and its contribution to farm economy in Indonesia

- A Case Study of Sukaresmi Village in Cianjur, West Java -

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1. Purpose

One of the main causes of deforestation in the tropics is the development of plantation agriculture. However, there exist serious problems such as low income for farmers and the wide-spread occurrence of insect pests due to the continuous growth of only one plantation crop.

Agroforestry is a combination of agriculture and forestry in space and time. Agroforestry can create more diverse, productive, profitable, healthy and sustainable land-use systems by growing trees (Westoby 1989). Also, the development of agroforestry in private forest contributed to not only increased forest cover and timber supply, but also farmers' household income (Iin 2005). Therefore, agroforestry systems in plantation land are necessary for reforestation in the future. This study investigated the relationship between agroforestry and farm economies on former plantation land, using Sukaresmi Village in the Cianjur district, West Java, Indonesia, as a case study.

2. Materials and methods

This study investigated the relationship between agroforestry and farm economies on former plantation land, using a case of Sukaresmi village. In order to clarify the relationship between agroforestry and farm economies, we conducted in-depth interviews with 30 farm households, who were doing agroforestry in the private forest from July to September 2012. The agroforestry management and farmers' income were surveyed. At the same time, we conducted interviews the village office and middleman, and sawmill, tea company. By these data, we analyzed contribution to farm economies and diversification of income from agroforestry.

3. Results

Sukaresmi village belongs to the Cianjur district located in West Java, Indonesia. According to the village office, the village is at an elevation of 700 to 800m, with an area of 1940 ha. As of 2011, the average annual temperature is from 20 to 30 °C, and the average annual rainfall is

3,075 mm. The population of the village was 3,756, with 1,971 males and 1,785 females.

The surveyed farmers possessed around 1.1 ha of private forest of agroforestry system on average. The total household income was Rp.23,321,606 per year (US\$1=Rp10,000) on average, and average income from agroforestry was Rp.6,497,466 per year, accounting for 28% of their total household income. In terms of the contribution of income from agroforestry to farmers' income, it can be categorized into three types of more than 50% (Type 1), 20-40% (Type 2), and less than 20% (Type 3). There were 11 households belonging to Type 1, meaning that their income highly depended on revenue generated from agroforestry, and they grew various crops different from the other types. There were 9 households of Type 2, many of whom grew tea (*camelia sinensis*). There were another 10 farm households of Type 3, many of whom grew woody plants mainly albizia (*Paraserianthes falcataria*).

4. Discussion

There are diversifications of income by agroforestry in Sukaresmi village, especially type1.

Farmers of Type1 grew various crops to prevent widespread occurrence of insect pests. On the other hand, type 2 and type 3 farmers grew a few crops or trees, because they have a job of non-farm. So they didn't have a time to plant grow up various species. And they didn't need to get a various income. But they grew various crops for self-consumption not cash income.

Also, trees were commonly harvested when they were 4 to 10 years old, and farmers obtained Rp.7,259,601 per harvest on average. Income from trees was not suitable as the main income source and did not contribute to daily households. However, trees had an important role in farmers' households, for example, which they need big cash for an emergency situation.

From the above results, it can be concluded that agroforestry contributed to farm economies, and the diversification of farmers' income in Sukaresmi village. However, some farmers who cultivated only tea wanted to introduce other crops and trees for the purpose of increasing the revenue, but they did not know how to plant or grow them. Currently, the management of private forest depends on each farmer. It is necessary to implement training in the agroforestry system for the farmers by the government or village office.

5. References

- 1) Westoby, Jack (1989) Introduction to world forestry
- 2) Ichwandi, Iin and Shinohara, Takeo and Darusman, Dudung and Nakama, Yuei (2005) Characteristics of Private Forest Management in Java, Indonesia: Two Case Studies. Journal of Forest Economics Vol.51(Nr.2): Page1-page12

Estimation of groundwater discharge in a sandy beach; An example of Fukiagehama, Kagoshima Prefecture, Japan

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1. Purpose.

Submarine groundwater discharge (SGD) is considered as an important source of dissolved material to the coastal zone¹⁾, recently. This study reveals a flow rate and volume of groundwater discharge in the Fukiagehama, Kagoshima, Prefecture, Japan were estimated.

2. Materials and methods

Study area;

Fukiagehama is located on the western coast of Kagoshima Prefecture, Kyushu, Japan (Fig.1).

Methods;

Water budget method (Fig.2) is applied to estimate the rate and volume of freshwater discharge in the Fukiagehama basin.

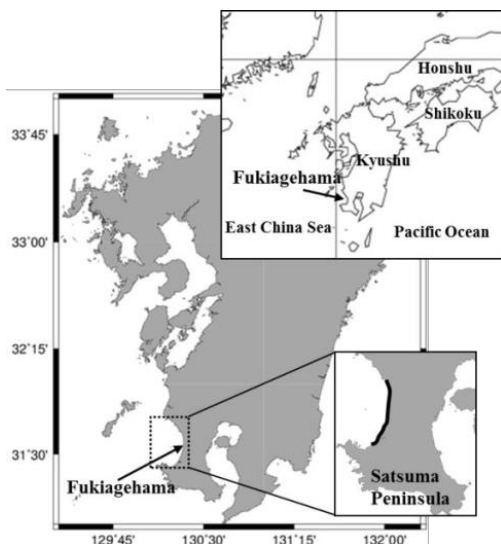


Fig.1 Study area.

$$P = R + G + E + \Delta S$$

P = Precipitation

R = River flow

G = Groundwater flow

E = Evapotranspiration { Thornthwaite method(E_T)
Hamon method(E_H)

ΔS = Change in water storage

Fig.2 Concept of water budget method.

3. Results

An average precipitation in the basin is approximately 2,200mm, and is greater than the Japan national annual average rainfall. It is seen based on the average value of the basin that the river flow is 29.7%, the groundwater flow is 36.6 to 37.8% of total amount of precipitation, and the rest is evaporated (Fig.3). Therefore, the river flow is 44%, the groundwater flow is 55.3 to 56% of total amount runoff (Table 1).

Table 1 Results of water budget method in the Fukiagehama basin.

Basin	Precipitation (mm/year)	Evapotranspiration(E_T) (mm/year)	Evapotranspiration(E_H) (mm/year)	Area×Precipitation ($\times 10^4 \text{m}^3/\text{year}$)	Area×Evapotranspiration(E_T) ($\times 10^4 \text{m}^3/\text{year}$)	Area×Evapotranspiration(E_H) ($\times 10^4 \text{m}^3/\text{year}$)
Eguchi River	2,155.2	893.0	875.9	4,935.3	1,646.5	1,601.1
Kamino River	2,194.2	918.4	898.3	21,678.6	7,350.6	7,107.1
O River	2,176.7	893.0	875.7	4,200.9	1,454.4	1,403.2
Nagayoshi River	2,239.4	925.6	904.4	11,286.4	3,726.9	3,613.1
Ono River	2,221.4	936.0	913.4	3,021.1	1,029.5	996.3
Izaku River	2,283.3	952.7	926.9	8,881.9	2,939.7	2,837.6
Manose River	2,339.7	967.9	938.9	87,108.8	29,304.5	28,163.2
Average	2,230.0	926.7	904.8	20,159.0	6,778.9	6,531.7

Basin	Runoff(E_T) ($\times 10^4 \text{m}^3/\text{year}$)	Runoff(E_H) ($\times 10^4 \text{m}^3/\text{year}$)	River flow ($\times 10^4 \text{m}^3/\text{year}$)	Groundwater flow(E_T) ($\times 10^4 \text{m}^3/\text{year}$)	Groundwater flow(E_H) ($\times 10^4 \text{m}^3/\text{year}$)
Eguchi River	3,288.8	3,334.3	1,009.2	2,279.7	2,325.1
Kamino River	14,328.1	14,571.6	4,288.9	10,039.2	10,282.7
O River	2,746.6	2,797.7	851.5	1,895.1	1,946.3
Nagayoshi River	7,559.5	7,673.3	2,207.5	5,352.0	5,465.7
Ono River	1,991.6	2,024.8	1,198.4	793.2	826.4
Izaku River	5,942.1	6,044.3	3,563.6	2,378.5	2,480.7
Manose River	57,804.3	58,945.7	41,777.3	16,027.0	17,168.4
Average	13,380.1	13,627.4	7,842.3	5,537.8	5,785.0

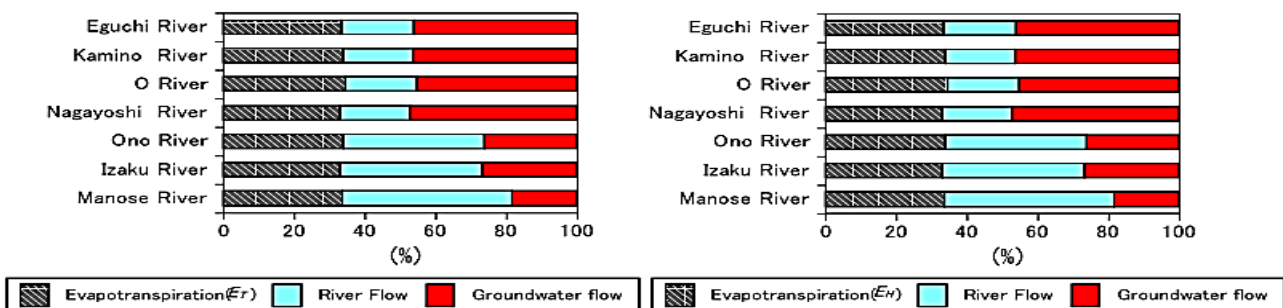


Fig.3 Individual ratio to total amount of precipitation.

4. Conclusions

It is clarified that quantity of groundwater can be large as $5.6 \times 10^7 \text{ m}^3/\text{year}$ due to the large amount of precipitation and soil condition (much volcanic debris known as SHIRASU) for which a permeability is high in general. Even though a macroscopic estimation using GIS, the groundwater flow is also important as a nutrient supply mechanism in the Fukiagehama basin.

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Effects of dietary fatty acids on cell differentiation and lipid accumulation of preadipocytes in cobia (*Rachycentron canadum* Linnaeus)

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1. Purpose

The present study established cobia preadipocytes culture system, and used the system to investigate the effects of dietary lipids or fatty acids on cobia preadipocytes proliferation, differentiation, lipid accumulation and adipogenesis genes expression. The aim was to explore the possibility modulate cobia lipid distribution and content through nutritional manipulation.

2. Materials and methods

In the present study, in addition to optimizing the culture medium for the differentiation of preadipocytes model, several studies evaluating the effects of fatty acids were carried out, including: comparisons of lipid accumulation and differentiation genes expression in adipocytes between in vivo and in vitro experimental settings, the effects of single fatty acid as well as multiple fatty acids on cobia preadipocytes differentiation genes CCAAT/enhancer binding protein- β (C/EBP β), peroxisome proliferator-activated receptors (PPARs), fatty acid binding protein1 (FABP1), fatty acid transport protein 2 (FATP2) and fatty acid synthase (FAS) expression.

3. Results

Cobia preadipocytes cultured in L-15 medium with 10% FBS at 28 °C was found to have a high proliferation potential leading to a monolayer confluence by day 18. The results of the in vitro and in vivo experiments were found to be compatible. Lipid accumulation was found to be greater in response to safflower oil, olive oil and palm oil treatments in both experiments than in fish oil and perilla oil treatments. At both molecular and cell levels, the effects of fatty acids on adipogenesis in vitro and in vivo systems showed similar trends. C/EBP β and PPARs expression increased during the early phase of differentiation and decreased at later stages of differentiation.

On the other hand expressions of FABP1, FATP2 and FAS were increased throughout the differentiation process. Linolenic acid (LNA) and docosahexaenoic acid (DHA) led to a lower accumulation of triglycerides (TG) in mature adipocytes than oleic acid (OA).

4. Discussion

We investigated the concordance between the gene expression profiles and key events of adipogenic development in the primary culture of cobia preadipocytes, including the induction of adipogenic differentiation by fatty acids (FAs), adipogenic gene expression, and lipid accumulation. The predominant FAs in fish oils behave differentially in shaping the adiposity of cobia adipocytes. This in turn suggests that variations in fatty acid profiles among different fish oils, which are the most important dietary lipid source in fish diets, could influence fat deposition in the flesh of the fish. Thus, adjusting the dietary fatty acid composition by using different fish oils or partially replacing them with plant oils might be strategically important when attempting to modify the flesh lipid content of cultured fish.

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Catch composition of tropical tuna associated with floating objects and free schools by purse seine in the Indian Ocean

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1. Purpose

Tuna is an important commercial fish, and there are 3 main species of tropical tunas namely skipjack (*Katsuwonus pelamis*), yellowfin (*Thunnus albacares*) and bigeye (*T. obesus*) which are distributed throughout the Pacific, Atlantic and Indian Oceans. In the Indian Ocean, there are a variety of fishing gears used by fishermen for catching tropical tuna and these include purse seine, longline, pole and line, and gillnet. The majority of catch are captured by the purse seine fishery (Pillai and Satheeshkumar, 2012). Fish aggregating devices (FADs) were first introduced into the tuna purse seine fishery as a way to attract pelagic fish such as tuna. In the last 30 years, FAD fishing has considerably increased the tuna yield for fishermen (Hallier, 1995), but the effect of FADs has also resulted in more by-catch of non-target species and small sized tuna. The hypothesis is that large sized tunas do not need to associate with floating objects as they usually form free schools, while small sized tunas aggregate on floating objects. The species composition on free schools and floating objects are different because of their behavior (Fréon and Dagorn, 2000). The purpose of this research is to compare between operation types with (I) the tuna catch per unit effort (CPUE) and (II) weight composition by fish size and species.

2. Materials and methods

Fishing data were obtained from Thai tuna purse seine logbooks that compose of fishing date, location; catch weight and operation types (free school, FAD, natural log or other floating object). The fishing grounds are located in the Western Indian Ocean between latitudes 10°S - 10°N and longitudes 45°E - 70°E.

The four operation types which are free school, FAD, natural log and other floating object were selected as the independent variables. The distribution of data will continue to be tested for the assumptions of statistical analyses (Zar, 1996). Average CPUEs and species composition of tunas were compared between each operation type. A probability of less than 0.05 was considered statistically significant. All statistical analyses were performed using the SPSS statistical program.

3. Results

Preliminary results indicate a total of 920 fishing operations were carried out by the Thai tuna purse seiners operating in the Western Indian Ocean. After a process of checking and verification through statistical analyses, only 876 data sets remained as the basis for this study. The catch data of tuna was separated by species and size. Skipjack tuna was sorted into 3 size classes by body weight: less than 1.8 kg, 1.8-3.4 kg and more than 3.4 kg. Bigeye and yellowfin tunas were divided into 2 size classes (≤ 10 kg and > 10 kg). The result of data distribution for each operation type showed that catch data did not have a normal distribution. Therefore, parametric statistics could not be tested on these data sets.

4. Discussion

Almost all of the fishing operations were conducted in the Western Indian Ocean. Some operations were carried out in the Central part of the Ocean, but all 44 of these data sets were omitted for the purpose of creating a homogeneous fishing ground. Pillai and Satheeshkumar (2012) further confirmed that the main fishing ground of Thai purse seiners is located in the Western Indian Ocean where most of the fish biomass is found. Therefore, data omission of the Central operations had little effect on the overall analysis.

Due to the characteristics of the data, assumptions for parametric statistics were not met thus, non-parametric statistics were used (Siegal, 1957). The average CPUEs and species composition of tunas will be compared among each operation type as the next steps for this study.

Tuna's importance in the world fish market is evident by such products as sashimi, loin and canned tuna; however, the fishery is not well managed in the Indian Ocean (Pillai and Satheeshkumar, 2012). The management on by-catch of under sized tunas is an issue of concern in the Indian Ocean, especially due to the purse seine fishery operating on associated sets. It is hoped that this research will provide some clarification to fishers about the issues of concern related to the tuna fishery associated with floating objects and introduce better techniques that may contribute to a more sustainable operations.

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Biological control of pathogenic root rot fungus

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1. Purpose

Pathogenic root rot fungus could infect many kinds of plants. In Kagoshima, which is one of sweet potato (Satsuma imo) main producing area in Japan, root rot has brought severe damage every year. Approaches to removing root rot have relied on using chemicals. However, the effect is insufficient. Biotechnological treatment to root rot has been investigated, such as using photosynthetic bacteria, actinomycetes, etc. In this study, biological control of pathogenic root rot fungus using photosynthetic bacteria, marine yeast, autochthonous bacteria was proposed. The antagonistic activities of different microorganisms and their combination were evaluated.

2. Method

Pathogenic fungus and autochthonous bacteria were isolated from the infected sweet potato farm (Kanoya City, Kagoshima, Japan) and maintained in the oatmeal medium (Hitoshi, 2009) at 25°C. One strain of photosynthetic bacteria (related to *Rhodopseudomonas faecalis.*), and 10 isolated autochthonous bacteria strains in this study were applied for a simplified disk placement method (Takanori et al., 1993) to evaluate their antagonistic activity. The root rot fungus was grown onto the oatmeal agar plates, and paper disks containing the culture supernatant were placed onto the fungus lawn. The plates were cultivated at 25°C and antagonistic activities were observed.

3. Result

Preliminary results showed that one strain of autochthonous bacteria, two combinations of autochthonous bacteria and photosynthetic bacteria showed antagonistic activity to root rot fungus. But limited inhibitory activity of photosynthetic bacteria only was observed. Practical use of the biocontrol agents in the infected field and details of the antagonistic mechanism are still under investigation.

4. Discussion

This study clarified the antagonistic characteristics of the autochthonous bacteria as potential agents to biocontrol the pathogenic root rot fungus. The achievement would be attractive from

the viewpoint of biological prevention against the root rot disease of cultivated sweet potato. The results of this study point out the antagonistic ability of the microorganism as potential material to treat pathogenic root rot fungus. The achievement of this study is attractive from the viewpoint of biological prevention and treatment to the root rot disease of cultivated sweet potato. As a next step, applicability of the antagonists to infected farms should be carefully investigated: impact of introduction of the antagonists to the farms on indigenous microbial community, possibility of the antagonists to dominate and maintain their populations in the infected farms, and their potential to expel the pathogen from infected plants and fields.

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Age and size effects on total and organic mercury concentrations in the white muscle of North Pacific albacore (*Thunnus alalunga*)

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1. Purpose

This study aimed to better understand age and size effects on mercury bioaccumulation in North Pacific albacore tuna (*Thunnus alalunga*), which is a commercially important fish stock.

2. Materials and methods

Using the methods of Chen *et al.* (2002) and Chen *et al.* (2011), we analyzed the concentrations of total mercury (THg) and organic mercury (OHg) in the white muscle tissues of 115 North Pacific albacore (fork lengths: 67–118 cm; body masses: 5.9–32.2 kg) collected from the catches of Taiwanese longline vessels from 2001 to 2006. The age of each fish specimen was estimated by investigating annual marks on a thin-sectioned sagittal otolith (Chen *et al.*, 2012). Using simple linear regression, we constructed the relationships between mercury (THg or OHg) concentration and age and between THg concentration and fish size (fork length or body mass).

3. Results

THg concentrations of the fish specimens ranged from 0.239 to 1.18 $\mu\text{g g}^{-1}$ wet weight (average $\pm$ SD: $0.445 \pm 0.148 \mu\text{g g}^{-1}$), and OHg concentrations ranged from 0.124 to 0.694 $\mu\text{g g}^{-1}$ wet weight (average $\pm$ SD: $0.283 \pm 0.091 \mu\text{g g}^{-1}$). Age estimates of the specimens ranged from 2⁺ to 13⁺ years. The simple linear regression between THg (or OHg) concentration and age indicated an average annual increment of 0.041 (95 CIs, 0.031 and 0.050) $\mu\text{g g}^{-1}$ for THg accumulation (**Fig. 1a**) and that of 0.025 (95 CIs, 0.020 and 0.031) $\mu\text{g g}^{-1}$ for OHg accumulation. Based on the simple linear regression between THg and fish size (fork length or body mass), we obtained an average increment of 0.008 (95 CIs, 0.006 and 0.010) $\mu\text{g g}^{-1}$ per 1-cm fork length and that of

0.017 (95 CIs, 0.013 and 0.021) $\mu\text{g g}^{-1}$ per 1-kg body mass (**Fig. 1b**). These results provide a better understanding of age and size effects on mercury contamination in North Pacific albacore.

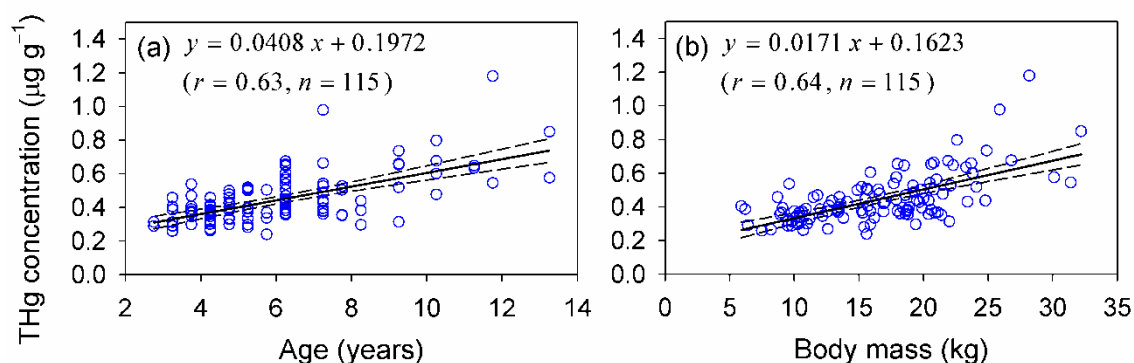


Fig. 1. The relationships between total mercury (THg) concentration and body mass and between THg concentration and age for North Pacific albacore (*Thunnus alalunga*). In each subplot, the solid line indicates the simple linear regression line, and the dashed lines show the 95% confidence intervals of the regression line.

4. Discussion

Based on the present result of OHg contamination (average: $0.283 \mu\text{g g}^{-1}$ wet weight) in North Pacific albacore, we suggest that the maximum weekly intake of albacore meat for a 60-kg person should be less than 339 g according to the provisional tolerable weekly intake (PTWI) of methylmercury (1.6 mg kg^{-1} body weight week^{-1}), set by FAO/WHO in 2003. Among the 10 fish of greater than or equal to 10 years of age, however, 9 fish (90%) had OHg concentrations greater than $0.283 \mu\text{g g}^{-1}$ wet weight, indicating the greater risk of methylmercury exposure from consuming meat of such old (or large) albacore. Except for the age and size effects, the higher mercury contamination of old (or large) albacore may be attributed to various biological (*e.g.* dietary shift, less growth dilution, and/or high bioaccumulation rate) and environmental (*e.g.* high mercury deposition) factors which need to be verified by further research.

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Warming induced inconsistencies in ocean acidification rates based on pH_{25} and $\text{pH}_{\text{insitu}}$

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Abstract: Ocean acidification is an unavoidable consequence of increased anthropogenic CO_2 , and even a small change in seawater pH may cause large impacts on oceanic ecosystems. In this study, acidification rates from two open access time-series studies are compared with that of six published time series studies. Conventionally, surface ocean is considered as under or close to air-sea CO_2 equilibrium. As a result, pH is expected to have an average decreasing rate of $-0.0016 \sim -0.0018 \text{ yr}^{-1}$ within the past 2-3 decades. The results show, however, although atmospheric CO_2 rises at similar rates globally, reported rates of changes in pH either measured at *in situ* temperature ($\text{pH}_{\text{insitu}}$) or at 25°C (pH_{25}) vary from $+0.00018 \sim -0.0024 \text{ yr}^{-1}$ to $-0.00071 \sim -0.0032 \text{ yr}^{-1}$, respectively. Instead of acidifying, in the South East Asia Time-Series (SEATS) study of the world's largest marginal sea—the South China Sea—the $\text{pH}_{\text{insitu}}$ changing rate is actually slightly increasing. Conversely, the pH_{25} at SEATS decreases at a rate 75% faster than expected under the air-sea CO_2 equilibrium. Such inconsistencies are consistent with the thermodynamics that $\text{pH}_{\text{insitu}}$ and pH_{25} time-series are incomparable as they change in anti-phase when seawater temperature changes. At the SEATS station, the seasonal change in $\text{pH}_{\text{insitu}}$ when temperature changes is 7-times of that expected under the air-sea CO_2 equilibrium. This is due to a positive feedback of warming on ocean acidification which implies that aquatic ecosystems suffering from ocean acidification could be affected earlier than expected when warming is considered along with increasing atmospheric CO_2 .

Reproductive seasonality of *C. annulus* (Mollusca: Gastropoda) in Hanazesaki, Kagoshima Prefecture, Japan

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1. Purpose

The objectives of this study are: 1) to determine the annual reproductive cycle of *Cypraea annulus* by detecting the change in gametogenic activity of their gonad through histological analysis; 2) to investigate sex ratio through gonad histology; and 3) compare shell lengths between two sexes.

2. Materials and Methods

Samples were collected randomly from intertidal rocky shore in Hanasezaki, Ibusuki, Kagoshima Prefecture, Japan (31°11'30.10"N, 130°30'29.70"E) from February 2011 to June 2012. Snails were fixed in 10% formalin after collection. Shell length was measured using digital caliper. Each shell was broken up to take out soft body. Gonads were removed and undergone standard histological procedures. Histological preparations were examined under the microscope. Reproductive stages were identified and classified as Stage 1 (initial maturity), Stage 2 (peak maturity) and Stage 3 (regression or post-gamete release).

3. Results

Male and female in the stage of peak maturity were found in almost all sampling periods, though their proportion varied seasonally. *C. annulus* seems to reproduce all year round (Figure 1). Female is usually larger than male in each season, however, the statistical significance was not found except in February 2011 and April 2012 (Figure 2). When all samples were pooled, shell length of females ($n=111$; $\text{mean}=23.5\pm 1.8\text{SD}$; $\text{range}=17.6\text{--}27.5\text{mm}$) was statistically larger than males ($n=84$; $\text{mean}=22.4\pm 1.9\text{SD}$; $16.7\text{--}26.4\text{mm}$) ($p < 0.0001$).

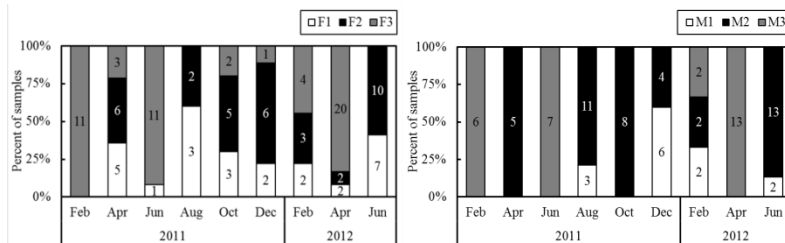


Figure 1. Percentage of reproductive stages of female and male *C. annulus* in each sampling period.

Number in each bar represents number of samples.

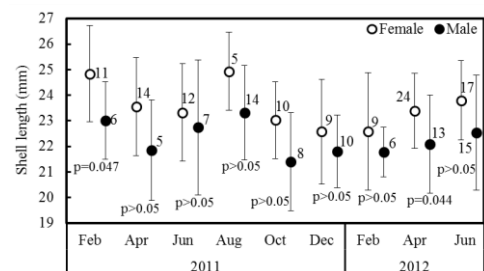


Figure 2. Shell length of female and male *C. annulus* (mean $\pm$ SD). p-values show the results of t-test on comparing shell size between two sexes. Number in each point indicates the number of samples.

4. Discussion

Sex of *C. annulus* was detected and the presence of various reproductive stages of females and males were identified through histological analysis. Difference between sexes was distinct after the three reproductive stages were categorized. The occurrence of more than one reproductive stage in most sampling periods suggests a continuous gametogenic activity of the reproductive cycle. Therefore, we can say that *C. annulus* reproduce all year round in tropical and sub-tropical areas as well as in temperate area with peaks from spring to early summer and from autumn to winter.

In this study, samples of females were more abundant than males and their size distributions indicated that females tend to be larger than males. This tendency was reported by Webber (1977) for species under genus *Cypraea*.

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Sponge diversity in the sea grass bed of Dongsha Island, Taiwan

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1. Purpose

Sponges are of great ecological importance in marine systems, but few studies have been conducted especially the habitat of sea grass bed. This study investigated sponge diversity at Dongsha Island which located at the west side of the remote Dongsha Atoll in Northern South China Sea.

2. Materials and methods

The sea grass beds around Dongsha Island are composed by 7 sea grass species and the meadow patches are nursery ground and shelters of numerous fish and invertebrates. Sponges were collected not only at southern, northern and northwest sea grass beds of the Island, but also from artificial concretes at south site.

3. Results

Seventeen sponge species were recorded which belongs to 12 genera, 8 families, and 6 orders. Among them, Poecioclereida is the most abundant Order. There are 2 species found only on artificial concretes, and some species were only found in a single sea grass site.

4. Discussion

The species compositions varied within 1 km in spatial scale. By comparison, sponge diversity in sea grass beds is lower than that of other marine habitats. Soft-sediment habitat and low current speed may result in a low diversity and high abundance sponge fauna in sea grass beds.

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Introduction of miso in Japan and manufacturing method

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1. Purpose

In Japan, miso has been eaten more than 1,300 years, and has contributed to the Japanese health. Miso is a typical seasoning of Japan as soy sauce, and also known as a functional food with rich nutrient. Nowadays soy sauce has been known and used all around the world, however miso has still poorly recognized. I would like to let you know the great feature of miso the base of Japanese food.

What is miso: Miso is a Japanese fermented food made from grain. The main raw material of miso is soybean which is mix with koji and salt then stand for long time to age.

About *Aspergills oryzae* and Koji: *Aspergillus oryzae* is a scientific name of one koji mold, used for miso, soy sauce, and sake fermentation. Koji is a koji mold *Aspergillus oryzae* grown on rice or barley or soybean.

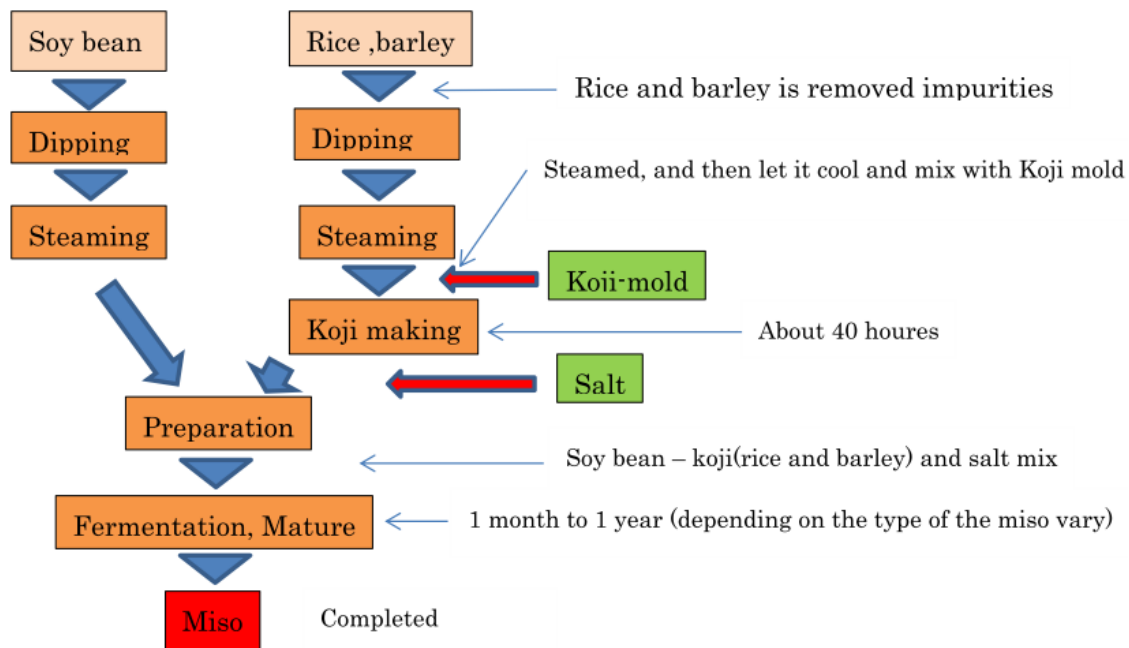
2. Benefits of miso

Nutrition: Soybean a main ingredient of miso contains a lot of high-quality protein. During fermentation and ripening, about 60% of soy protein are converted to water soluble form and about 30% to amino acid. Thus, eating miso is better in digestion and absorption of proteins and amino acids than eating soybean itself. Also, miso is rich in carbohydrate, lipid, dietary fiber, vitamins, minerals, and other nutrients.

Decrease action of functionality.

- [1]. Lowering effect of cholesterol.
- [2]. Cancer prevention action.
- [3]. Gastric ulcer prevention effect.
- [4]. Anti-aging action.
- [5]. Removal action of radioactive material.

3. Miso production flow chart



5. My research goals

By controlling the proliferation of microorganisms to establish high quality miso and soy sauce production methods in the stable.

6. References

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Response of fishers toward application of fishery management and development initiatives.

Sub-title: A comparative study on responses of fishers of Tuvalu, Japan and Malaysia.

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1. Purpose

Current fisheries management depend on stock assessment to estimate population parameters of the focal species from the age or length structure of past catches, biomass of past catches, past fishing effort, and fishery-independent surveys. Fisheries scientists recommend potential management action based on these estimates to fisheries managers for final decision (Botsford, 1997). This is actually a scientific approach to fishery management. However, Pomeroy, 1995, emphasised that, fishery experts recognised that the underlying causes of fisheries resources over-exploitation and coastal environmental degradation are often of social, economic, institutional and/or political origins. He further claims that the concerns of fisheries management should address the relationship of fisheries resources to human welfare, present and future and therefore people should be the main focus. These are two of the approaches toward fishery management, but there are also recent initiatives such as Ecosystem Approach Fishery Management which is currently adopted by certain countries.

This study is basically centred on the approach which people are considered as important factors in fishery management. Since legal documents such fishery acts and regulations are management measures commonly used in every country to control fishing activities, the study is basically focus on fishers perceptions toward application of these management and development measures. In fact the main objective of the study is:

- a) To compare and contrast the responses and perceptions of fishers toward application of fishery management and development initiatives.

The study basically target to identify and compare the responses of fishers in Japan, Malaysia and Tuvalu toward fishery management and development initiatives in their respective coastal fishery waters. Prior to the acquirement of information from fishers, the study feels that

understanding of the fishery management systems of these respective study sites is vitally important. This lead to the sub-objective of the research which is:

b) To identify and compare the existing fishery management systems of Japan, Malaysia and Tuvalu.

2. Materials and Methods

Data on fishers' responses will be collected using questionnaires. Questionnaires will be created to investigate the opinions and perceptions of fishers towards fishery management measures such as fishery regulations and closed areas, and fishery development initiatives such as a set-net fishery.

The analysis of fishery management systems will be carried out by reviewing legal documents such fishery acts, fishery regulations and other legally-bind documents that are responsible for fishery management in these countries.

3. Results

Data have been collected in one of the study sites (Malaysia) and are still in the analysis process, while other sites still await the planned dates. The analysis of fishery management systems are still underway and yet to be completed.

4. Discussion

It is believed that any management and development measure impose on marine resources will directly affect fishermen socially and economically. The level of compliance or the effectiveness of any fisheries management tool and the successfulness of any fishery development can depend on the reaction of fishermen.

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Methodological studies in data limited fisheries: Development of new methodology to calculate gillnet selectivity curve and estimation of population length distribution of pink salmon (*Oncorhynchus gorbuscha*)

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1. Purpose

Gillnets are widely used for the harvest of fish (Ries and Pawson, 1992; Carlson, 2003), because gillnets are highly selective for fish of certain size (Hamely, 1975), require little investment and equipment and are effective in catching widely scattered fish (Ries and Pawson, 1992), and the ease of use and possibility to be set at any depth and in areas with difficult bottom condition as well (Hovgard and Lassen, 2000). Knowledge of size selection of gillnets is vital to the proper management of fisheries. It is essential to estimate population length distribution, set mesh size regulation and when information of the growth, Fecundity and mortality of the species is available models can be used to determine strategy for optimizing sustainable yield of the fishery (Gulland, 1983; Miller 1992; Miller and Host, 1997; and Huse et al,2000).Furthermore, when gillnet size selectivity of successive year is estimated, could be used to monitor effect of fishing in data-limited fisheries of developing countries where total allowable catch (TAC) of the fisheries is unknown. Several methods have been developed to estimate gillnet selectivity (Ishida, 1965, Kitahara, 1968, Kawamura, 1972, Matsuoka and Kawamura, 1992), using indirect method, determined the gillnet selectivity by comparing the length frequency distribution of fish caught by gillnets of different mesh sizes. The objective of the present study is to develop a new methodology to estimate gillnet selectivity curve based on methodology developed by Ishida's 1965 and Matsuoka and Kawamura, 1992.

2. Methodology

Data necessary for calculation of mesh selectivities were obtained from drift gillnets used in fishery independent survey of Salmonid population in North West Pacific Ocean (155°E latitude and between 36°N and 46°N latitudes),the research line of the Graduate School of Fisheries Sciences and Faculty of Fisheries, Hokkaido University, Japan. Pink salmon *Oncorhynchus gorbuscha* were collected with drift gillnet consisting panels of 21 different mesh size. Stretched mesh sizes ranged from 19mm to 157mm categorized in to three groups: the F-Gear and are also called special mesh (19mm, 22mm, 25mm, 29mm, 33mm, 37mm, and 42mm), the C-Gear,

called non-selective varied research mesh (48mm, 55mm, 63mm, 72mm, 82mm, 93mm, 106mm, 121mm, 138mm, 157mm, 112mm, 118mm), and A- Gear also called commercial mesh (115mm, 121mm) were used (Takagi,1975). The drift gillnet is composed of 30 tans of C-Gear gillnet, 12 tans of A-Gear gillnet, and seven tans of F-Gear gillnet, total of 49 tans. Each tan is 50m long, the research drift gillnet is therefore 2450m long.

Experimental fishing have been conducted, by Oshoro Maru and Hokusei Maru research vessels of Hokkaido University, Fisheries Faculty, Japan, between May and June every year since 1953 (Hokkaido University,2010: Hoshi 2010).In the present Study, the data obtained from the survey conducted by Oshor Maru during May 2009 is used. Drift gillnets were set in the evening, soaked overnight, and retrieved in the following morning. Pink salmon captured were measured in fork length (mm).

Gillnet selectivity curve and fish length at maximum selectivity will be estimated using the new methodology. This methodology will be developed based on Ishida method 1965 and Matsuoka and Kawamura 1992 KM method. Least square difference and solver function will be applied. The method will be also compared if it could give the same result when developed from Kithahara's method 1968. and KM method.

3. Discussion

The research is currently in progress. New methodology to calculate gillnet selectivity curve will be developed. Gillnet selectivity curve and population length distribution of pink salmon will be estimated. The outcomes of this research will improve the gillnet selectivity curve estimated by Ishida in 1965.This method will give best fit model and improve the flatted head of the selectivity curve. Moreover, this method is cost effective and essential especially to the management of data limited fisheries of developing countries to monitor the effect of fishing by comparing the population length distribution of successive years and develop mesh size regulation to ensure development and the long term sustainability of the fisheries.

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Climate effects on temporal variation in abundance and distribution of the demersal fish assemblage in the Tsushima Warm Current region of the Japan Sea

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1. Purpose

To understand the climate effects on the temporal variation in abundance and geographical distribution of demersal fishes at both interannual and decadal scales, and how well can the ecological and life history traits explain the sensitivity of species responses.

2. Material and methods

We analyzed the single-trawler fishery data (1972-2002) in the Tsushima Warm Current region of the Japan Sea (Tian et al., 2009; 2011). We first describe the environmental changes of the Tsushima Current region of Japan Sea through both local and basin-scale environmental indicators. We then investigate whether the changes in distribution and abundance are significant at interannual (1972-2002) and decadal scale (between the cold (1977-1988) and warm (1989-2002) period). Finally, we check how well the geographic affinity and life history traits can explain the sensitivity of species responses by logistic regression.

3. Results

Our results indicate that changes in distribution as response to interannual climate variability are best explained by geographical affinity, as most cold-water species exhibit a poleward shift. For species with changes in distribution at both interannual and decadal scales, the marginal significance in asymptotic length suggests large species are more likely to demonstrate distributional changes. But none of the traits examined in this study are able to explain the change in abundance at interannual or decadal scale.

4. Discussion

For the change in distribution at interannual scale, the difference in sensitivity may lie in the physiological adaptation of fish. The cold water species usually have subarctic origin and narrower thermal tolerance limit, which make them vulnerable to environmental change (Pörtner and Peck, 2010). None of the factors we examined can explain the decadal patterns, suggesting other factors may need to be considered. Likewise, none of the life history and ecological trait variables can explain the change of abundance at interannual, decadal scale or combined. This suggests that the nonlinearity in response may be important for understanding the dynamics of exploited species in a changing environment.

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Population structure of Limpid-wing flyingfish (*Cheilopogon unicolor*) in the north-western Pacific inferred from mitochondrial CO I genes

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1. Purpose

Limpid-wing flyingfish (*Cheilopogon unicolor*), a pelagic migratory fish is one of the dominant flyingfishes in waters around Taiwan. However, *C. unicolor* was not found in northern Philippines and southern Japan. There are similar species *Cheilopogon furcatus* in the Philippines and *Cypselurus antoncichi* in Japan. In case of misleading denomination of *C. unicolor* in three nations, it would confuse fishery and research. Therefore, this study analyzed the population structure of *C. unicolor* in northern-west Pacific Ocean.

2. Materials and methods

We collected 44 specimens in 10 localities and used mitochondrial COI fragment (634 bp) to analyze the population structure.

3. Results

The total hapotype diversity was 0.899, nucleotide diversity 0.383%, the average genetic distance 0.59% and the fixation index between 0.042-0.19. The Neighbor-Joining tree shows no apparent separating lineages. The gene flow of *C. unicolor* in northern-west Pacific Ocean was high and belonged to one stock.

4. Discussion

The study suggests that the three species are the same species. *Cheilopogon furcatus* is probably a misidentification in Philippines and *Cypselurus antoncichi* is junior synonym in Japan. Considering the migratory feature of the species, strengthening of cooperation between Japan and Taiwan is needed.

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Development of the LED fishing light based on the visual sensitivity of fish

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1. Purpose

Artificial light is widely used in Japanese fisheries for the purpose of aggregating target animals to fishing vessels. Recently, following the high power LED development, energy saving LED fishing light has been successfully installed in the pacific saury stick-held dip net and other types of fisheries. The use of LED fishing light is thought to spread to various light fisheries as a consequence of the high price of fuel. Therefore, it is essential to compare the efficiencies of conventional lights, such as halogen and incandescent, and the newly developed LED lights for optimum installation of LED fishing light into commercial fisheries. Light emitted from the source travels in the water and is perceived through the vision of target animals; hence, both the water properties and visual sensitivity to the light spectrum are the factors that have to be incorporated into numerical simulations. In this paper, we introduce our recent studies on the measurements of optical transparency of water and visual sensitivity of fish.



Fig. 1

A fishing vessel equips LED fishing lights. This vessel operates stick held dip net (“bou-uke-ami” in Japanese name) in Kagoshima and main captive species is Japanese anchovy *Engraulis japonicus*, round herring *Etrumeus teres*, Banded blue sprat *Spratelloides gracilis*, and Jack mackerel *Trachurus japonicus*.

2. Materials and Methods

2-1. Basics of light propagation and perception sequences in the sea

Figure 1 illustrates the sequence of underwater light propagation and perception during light fishing operation. Several types of the light source are used in fishing operation and those have different spectral component. Light intensity is attenuated while traveling in water. The Beer-Lambert Law equation (eq. 1) shows the relation between the traveling distance (z) and light intensity (I).

$$I_z = I_0 e^{-cz} \quad (\text{eq. 1})$$

Attenuation rate is expressed by c (defined as the attenuation coefficient) in the equation. It should be noted that c is largely different by the wavelength of light and turbidity of water. In most of the

case, the lowest c value is obtained at blue-green color light, around 490 - 500 nm wave length while much higher toward shorter (to purple) and longer (to red) wave length.

On the other hand, visual sensitivity of fish is different from our eye. Fish retina possesses 2 kinds of photoreceptor, cone and rod cells. Since light fishing is operated in the night, rod cell which is functional under dim light condition may closely relate to aggregation behavior to the light. Therefore, sensitivity of the rod cell is the essential knowledge to discuss fish behavior and optimum design of the LED wavelength.

2-2. Measurement of light attenuation coefficient in each wavelength

To obtain the attenuation rate of light during its propagation in the sea, photon flux density (PFD, from 380 - 700 nm, $\mu\text{mol}\cdot\text{nm}^{-1}\cdot\text{s}^{-1}$) of sun light is measured by a spectrophotometer at various water depth in daytime. Then, reduction coefficient (c) is obtained by fitting the measured PFD values to the eq. 1 in each wavelength.

2-3. Visual sensitivity of fish

An electrophysiological method, recording the electroretinogram (ERG) is used to obtain the sensitivity curve of rod cell. Fish exposed to the dark is immobilized and electrical potentials in response to the short light pulse, at various intensities and wavelength, were recorded. Using the self-developed analysis protocol, visual thresholds and sensitivity curves were obtained.

3. Results and Discussion

3-1. Properties of attenuation coefficient

We have been estimating the attenuation coefficient in Kagoshima bay and other areas to know the seasonal changes of water properties and to know the difference in coastal and oceanic waters. So far, it is clearly shown that attenuation coefficient c tend to show higher value in coastal water particularly during summer, however, the lowest c is always seen at 490-500 nm wavelength. Such obtained coefficient will be used for the simulation on the light distribution around the various type of fishing light to compare the efficiencies of conventional halogen, incandescent and LED light.

3-2. Visual sensitivity properties

Our laboratory has been conducting the ERG recording in Jack mackerel *Trachurus japonicus*, Pacific saury *Cololabis saira*, and Japanese anchovy *Engraulis japonicus*. So far, Visual sensitivity curves of rod cell in each species show slightly different property, however the highest sensitivities appeared around the range of 500 nm. Our results suggest that fish vision particularly the rod cell sensitivity is well adapted to the underwater light propagation properties.

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Dynamics of subtropical zooplankton is steady-state? As an evidence for seasonal changes in population structure and depth distribution of two subtropical copepods.

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1. Purpose

Copepods are the most predominant group of zooplankton community and contribute fishery production in the world's oceans (Mauclaine 1998). There is increasing information that they have great impacts on trophodynamics and biogeochemical cycles in marine ecosystems through their diel and seasonal dynamics (Kobari et al. 2003, 2008, 2010). While subtropical zooplankton community has been long considered to show no or less seasonality (e.g. McGowan and Walker 1979, 1985), some subtropical copepods are suggested to adapt their life cycles to seasonal food availability (Shimode et al. 2009, 2012a, 2012b). In the present study, we investigate the life cycle strategies of two subtropical copepods (*Rhincalanus nasutus* and *R. rostrifrons*) to food availability through seasonal zooplankton samplings and oceanographic observations.

2. Materials and Methods

Oceanographic observations and zooplankton samplings were carried out during four cruises (November, February, April and July) on R/V Mirai at S1 (30°N, 145°E) in North Pacific Ocean. Copepods were collected at 8 depth intervals in 0-1000 m with an Intelligent Operative Net Sampling System (IONESS: mesh size 0.33 mm) during the day and night. *R. nasutus* and *R. rostrifrons* were identified into 6 copepodite stages and enumerated under a dissecting microscope. Chlorophyll *a* concentration and primary production were also determined from water samples.

3. Results

Primary production and chlorophyll *a* concentration showed seasonal changes with a maximum in February. *R. nasutus* occurred more abundantly in the water column than *R. rostrifrons*. While both two species appeared throughout the four seasons, *R. nasutus* and *R. rostrifrons* reached a maximum abundance in May and November, respectively. Adult males and females were abundant in February. Both copepods concentrated in the surface layers above 200 m during February, but descended into the mesopelagic layers below 300 m in the other seasons. In particular, both copepods disappeared from the surface layer in November. The predominant stage was copepodite stage 5 and adult female in the mesopelagic layers.

4. Discussion

Adult males and females appeared more abundantly in February when food availability estimated from primary production, chlorophyll *a* and unicellular plankton biomass was the highest. They

produced the mesopelagic stocks in the other seasons. These present findings support the previous results that they adapt their life cycles (i.e., reproduction and recruitment) to seasonal fluctuations of food availability (Shimode et al. 2012b). These results suggest that dynamics of subtropical zooplankton are not steady-state.

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Nucleic acid ratios can be a proxy for egg production rate of various marine copepods?

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1. Purpose

Copepods are the most numerous metazoan on the planet and produce huge biomass in marine ecosystems (e.g., Mauchline 1998). They integrate energy from the microbial and grazing food webs and transfer it into fishery production. Thereby, considerable efforts have been expended in measuring secondary productivity over the past few decades. Egg production is a common approach to quantify secondary production, while this method can be applied only for egg-laying animals and the estimates are suggested to involve the large and variable errors (Hirst and McKinnon 2001). On the other hand, biochemical approach using nucleic acids and protein has been proposed to evaluate physiological process for various zooplankton (see Kobari 2010). The biochemical approach would give some information on the large and variable errors in egg production rates, however, there is little comparison between the two estimates by egg production method and biochemical approach.

Here, we compare egg production rates to individual contents and the ratios of nucleic acids and protein for marine copepods. From these results, we consider the large and variable errors in egg production rates using the biochemical proxies.

2. Materials and methods

Calanus marshallae, *C. pacificus* and *C. sinicus* were selected as model species since *Calanus* species are known to be important for secondary production in the world's oceans. Copepod samplings were carried out during cruises on R/V Elakha off Oregon coast, USA (for *C. marshallae* and *C. pacificus*) and T/S Nansei-Maru in Kagoshima Bay, Japan (for *C. sinicus*). Copepods were collected from near bottom to sea surface using a Bongo net and North Pacific Standard net (mesh size 0.33 mm) with a large cod end. Adult females were identified into species, development stages and sex (i.e., adult female) under a dissecting microscope. Each specimen was transferred into a 20 mL glass dish and incubated during 36 hours under dark condition and ambient temperature. After counting eggs and nauplii, each specimen was immediately preserved under -80°C for nucleic acids and protein analyses.

RNA and DNA were measured following the microplate fluorescent assay (MFA) developed by Wagner et al. (1998) and Kobari et al. (2013). To define the body mass of each specimen, aliquot of the sample homogenate was used for protein determinations according to the method of Bradford (1976).

3. Results

C. sinicus showed the lowest egg production rate (0-26 eggs day⁻¹) while egg production ratio was comparable for *C. marshallae* (0-89 eggs day⁻¹) and *C. pacificus* (0-72 eggs day⁻¹). The three species

showed no significant correlation of the egg production rates to the individual content of DNA, RNA and protein because the animals laying no egg revealed that the individual contents were variable. Even though the animals laying no egg were excluded, the three copepods indicated no or less significant correlation of the egg production rates to the individual RNA, DNA and protein contents. Egg production rates of the three copepod species demonstrated no significant correlation to the RNA:DNA and RNA:protein ratios.

4. Discussion

While significant correlation of egg production to nucleic acid ratios was reported for *Paracalanus* sp. (Nakata et al. 1994) and *Acartia bifilosa* (Holmborn et al. 2008), such correlation was not found for the three *Calanus* species. According to Hirst and McKinnon (2001), egg production does not always represent growth of many copepod species, since adult body weight is variable whether or not eggs are exuded. Non-steady-state body weight is well known for *Calanus* species due to the lipid consumption for gonad maturation and egg production (Plourde and Runge 1993). Considering that the biochemical approach using nucleic acids and protein (i.e., protein synthesis) does not represent lipid consumption, egg production rate for *Calanus* species might not be associated with RNA:DNA and RNA:protein ratios.

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Analysis of trophic dynamics in zooplankton community using stable isotope ratios

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1. Purpose

It is well known that a wide range of planktonic organisms can be supported even in the limited resources of pelagic marine ecosystem as one of the paradox of the plankton (Hutchinson, 1961). Since the competitive exclusion is principle for the other ecosystems (Hardin, 1960), considerable efforts have been expended to clarify the paradox in marine ecosystem over the half century. However, we have still limited knowledge. Recently, stable isotope technique has been used for understanding complicated food webs in many ecosystems (Hecky and Hesslein, 1995; Marguillier et al., 1997). Indeed, Aita et al. (2011) suggest that stable technique can be used to elucidate general patterns in planktonic ecosystems and biogeochemical cycles.

In the present study, we compare the seasonal changes in carbon and nitrogen stable isotope ratios of zooplankton community between the two different sites and among three depth ranges to consider trophic dynamics of zooplankton community in different environment fluctuations.

2. Materials and Methods

Oceanographic observations and zooplankton samplings were carried out during four cruises (November, February, April and July) on R/V Mirai at K2 (47°N, 160°E) and S1 (30°N, 145°E) in North Pacific Ocean. Zooplankton samples were collected at 8 depth intervals in 0-1000 m with an Intelligent Operative Net Sampling System (IONESS: mesh size 0.33 mm) during the day and night. The aliquots were immediately frozen at -20°C. Metazoans were identified into species or genus levels under a dissecting microscope, and dried at 60°C for 24 hours. After eliminating their lipids, carbon and nitrogen isotopic ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) of the specimens were determined using an elemental analyzer/isotope-ratio mass spectrometer.

3. Results

In the surface layers above 150 m, the $\delta^{15}\text{N}$ values of carnivores were higher than those of particle-feeders throughout the seasons at K2, while such pattern was not clear for zooplankton community at S1. While the lower $\delta^{15}\text{N}$ values were found for the zooplankton community at S1 compared with those at K2, they showed the seasonal patterns with the wider range of the $\delta^{13}\text{C}$ values and the higher $\delta^{15}\text{N}$ values from autumn to spring at K2 and spring to summer at S1. Although the seasonal patterns were diminished for the zooplankton communities in the upper (150 to 500 m) and lower mesopelagic layers (500 to 1000 m), the $\delta^{13}\text{C}$ values were within the range of those of the surface zooplankton community. The $\delta^{15}\text{N}$ values were higher for the zooplankton community in the deeper layers.

4. Discussion

Since the $\delta^{15}\text{N}$ values were lower for nitrogen fixers like cyanobacteria compared with those for the other phytoplankton (Carpenter, E. J. et al. 1997), the lower $\delta^{15}\text{N}$ values mean that the surface zooplankton community at S1 consume the nitrogen fixers or protozoans feeding on them as a food resources. Considering with the enrichment of $3\pm 1\text{‰}$ of $\delta^{15}\text{N}$ (Minagawa and Wada, 1984) and 1‰ of $\delta^{13}\text{C}$ (Rau et al., 1983) per one trophic level, the higher $\delta^{15}\text{N}$ and the wider $\delta^{13}\text{C}$ range with the seasonal transition would indicate that surface particle-feeders consume not only phytoplankton but also protozoans from autumn to spring at K2 and spring to summer at S1 as shown in Doi et al (2010). While the seasonal pattern was diminished for the mesopelagic zooplankton community, the higher $\delta^{15}\text{N}$ with depth suggest that the mesopelagic particle-feeders and carnivores consume food items (e.g., live or dead animals, fecal pellets or exuvia) facilitated by from carnivores. The present findings suggest that carbon and nitrogen stable isotope ratios are useful for understanding trophic dynamics of not only epipelagic but also mesopelagic zooplankton community.

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Effect of towing time for a bottom trawl on the freshness of shrimps

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1. Purpose

Shrimps are one of the main marine species caught by bottom trawls. The price of shrimps may increase if the shrimps are as fresh as possible, resulting in an increase in the income of the fishermen. Therefore, in this study, I clarified the effect of towing time for a bottom trawl on the freshness of shrimps.

2. Materials and methods

Between June 2012 and June 2013, experimental trawling was performed by our training vessel Nansei-maru in Tarumizu, Kagoshima Bay. A data storage tag (DST milli-F) that recorded temperature and depth was attached at the center of the head rope and ground rope, and measured every second. Both bottom towing of grounding (10 min) and extended towing of non-grounding (0.5, 1.0, and 2.8 h) were performed. The length of the warp for bottom towing of grounding and extended towing of non-grounding was 900 m, and 400 m, respectively, and the towing speed was 2.0 knots. The deep-water mud shrimps *Solenocera melantho* were kept on ice immediately after they were caught and were frozen using dry ice after 0, 1, 6, and 12 h in order to analyze ATP related compounds (ATP, ADP, AMP, IMP, HxR and Hx).

The flow speed in the trawl net during grounding and non-grounding was measured by a flume tank experiment with a 1/8th scale model net. The flow speed of the flume tank was 0.46-0.77 m/s, which is equivalent to the prototype being towed at 1.5–2.5 knots in the sea.

3. Results

The model experiment showed that the flow speed in the net from the mouth of the net to the cod end during grounding and non-grounding were 0.59 m/s at experimental setting flow speed of 0.62 m/s. Thus, the flow speed in the net of grounding and non-grounding was the almost same.

Table 1 and 2 show the results of the analysis of the ATP related compounds and K value in specimens caught by bottom towing of grounding and bottom towing of grounding plus extended towing of non-grounding. The result of the analysis of the ATP related compounds showed that the amount of ATP immediately after being caught was 0.12 $\mu\text{mol/g}$ in specimens caught by bottom towing of grounding and 0.69 $\mu\text{mol/g}$ in those caught by bottom towing of grounding plus extra towing of non-grounding (2.8 h). Thus it was assumed that the effect of the amount of ATP on the specimens did not differ with towing time between 11 minutes and 2.8 hours. In addition, HxR and Hx were not detected in all of the samples. Therefore, K value was 0% in the all samples.

Table 1.

ATP related compounds and K value of bottom towing of grounding, and bottom towing of grounding plus extended towing of non-grounding.

Bottom towing (min)	Extended towing (h)	ATP related compounds ($\mu\text{mol/g}$)						K value (%)
		ATP	ADP	AMP	IMP	HxR	Hx	
11	0	0.12	0.77	3.47	1.85	-	-	0.0
15	2.8	0.69	0.94	3.03	1.62	-	-	0.0

Table 2.

ATP related compounds and K value of bottom towing of grounding, and bottom towing of grounding plus extended towing of non-grounding.

Bottom towing (min)	Extended towing (h)	ATP related compounds ($\mu\text{mol/g}$)						K value (%)
		ATP	ADP	AMP	IMP	HxR	Hx	
11	0	0.05	0.55	2.81	0.22	-	-	0.0
11	0.6	0.03	0.59	2.75	0.52	-	-	0.0
11	1.1	0.25	0.67	3.12	0.19	-	-	0.0

4. Discussion

Because HxR and Hx were not detected in all of the samples of the deep-water mud shrimps, The ATP related compounds did not disintegrate. The above mentioned results showed that the towing time until 2.8 h did not have effect on the freshness of deep-water mud shrimp.

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Evaluation of the freshness between catching and landing for stick held dip net fishery

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1. Purpose

Japanese anchovy *Engraulis japonicus*, round herring *Etrumeus teres* are main target species of stick held dip net fishery in Kagoshima. The fish landed using stick held dip net fishery are processed or used as bait of aquaculture. A solution for the problem such as reduction of price may be to increase the consumption of fish as fresh food. However, it is difficult to implement this solution because of variation in the freshness of the catch.

In this study, we evaluated the freshness of round herring and Japanese anchovy within 6 hours of catching them by using stick held dip net fishing.

2. Materials and method

We boarded a fishing boat of stick held dip net fishery on June 13, 2012 and March 8, 2013, and recorded the fishing process from catch up to landing, water temperature of the fishing grounds, and changes in the water temperature of the fish hold. We collected 10 samples each of anchovies and round herrings every hour for 6 hours immediately after catching the fish.

ATP related compounds (ATP, ADP, AMP, IMP, HxR, and Hx) were obtained from the samples, and freshness was evaluated by measuring the concentrations of ATP related compounds in the samples. K value was calculated using the concentrations of ATP, ADP, AMP, IMP, HxR, and Hx. The stiffness index was measured to examine the progress of rigor mortis.

3. Results and Discussion

Figs. 1 and 2 show the concentrations of ATP related compounds extracted from round herring, and Japanese anchovy, respectively. Fig. 3 shows the K value of round herring and Japanese anchovy, and Fig. 4 shows the stiffness index of round herring and Japanese anchovy.

It was found that the concentration of IMP was high immediately after the fish were caught, and IMP had not been converted to HxR in round herring (Fig. 1). The concentrations of HxR was higher in Japanese anchovy than in round herring, and they had not been converted to Hx (Fig. 2). The K value for round herring was less than 10%; the K value for Japanese anchovy fluctuated between 29% and 42% (Fig. 3). Fig. 4 shows that rigor mortis was initiated immediately after the catch for round herrings and was started 2 hours after catch for Japanese anchovy.

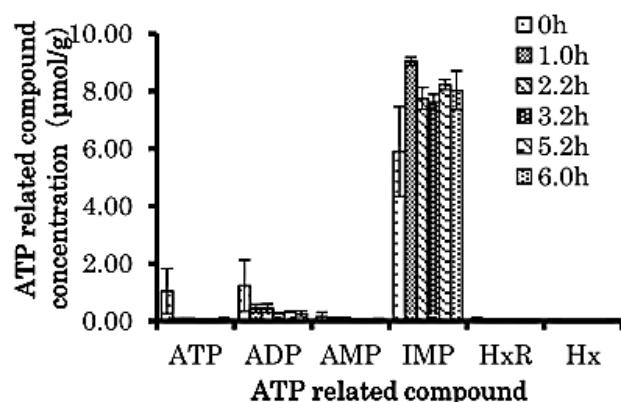


Fig.1 Relation between the ATP related compounds of round herring, and the reserve time after catching.

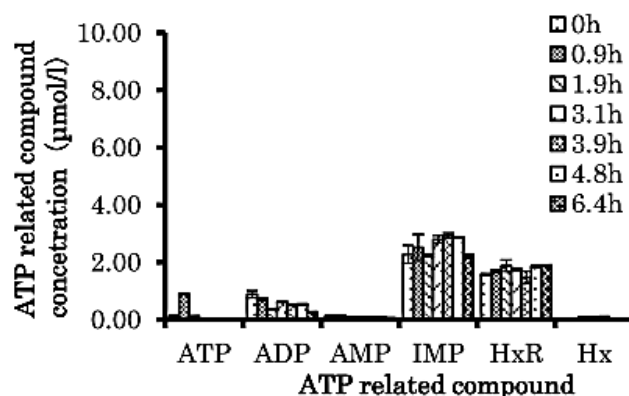


Fig.2 Relation between the ATP related compounds of Japanese anchovy, and the reserve time after catching.

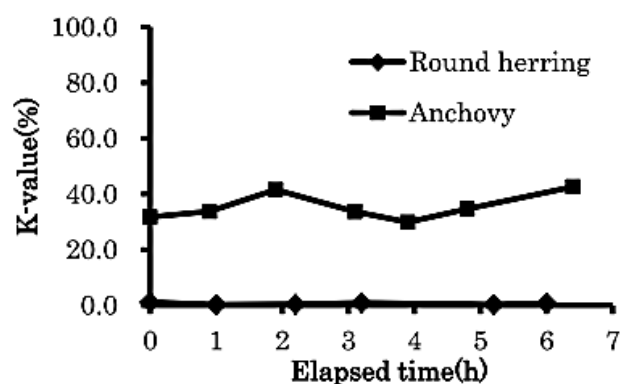


Fig.3 Changes of K value of round herring and Japanese anchovy.

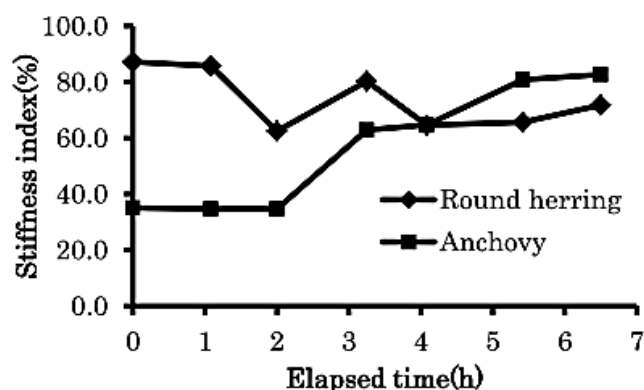


Fig.4 Changes in stiffness index of round herring and Japanese anchovy.

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Analysis of turning behavior of reared Pacific Bluefin tuna in an aquaculture net cage

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1. Purpose

Aquaculture of Pacific bluefin tuna become remarkable development although the aquaculture has a problem about high mortality of the juvenile reared fish (Ishibashi, 2006). 50% of mortality of reared fish occurs after release into an aquaculture net cage. The reducing number of the reared fish go on about for one year from the releasing. The firmer of the aquaculture commonly said that the dying of reared fish are always found at morning. Mortality of bluefin tuna was caused by collision with the net of the cage that fatally damaged the bones of the vertebral columns and the parasphenoid (Miyashita, et al, 2000). High mortality of Pacific bluefin tuna in night-time caused by poor scotopic vision (Ishibashi, et al, 2009). Therefore, we have to investigate the behavior of reared fish from night to day. The main of goal of this study is to obtain scientific understanding of turning behavior for improving their mortality ratio using a dead reckoning technique in tuna aquaculture net cage.

2. Materials and methods

This study was carried out at an aquaculture net cage located offshore (80 m deep) of Kochi, Japan. The tagged fish was angled from the cage and surgically attached data loggers (W190-PD3GT : Little Leonardo, 21 mm diameter, 117 mm in length, weight 75 g in air; DST Comp-Tilt : Star-Oddi, 15 mm diameter, 46 mm in length, weight 19 g in air) near its dorsal fin for measuring its swimming behavior, the swimming speed, swimming depth, heading at 1 s intervals. The three-dimensional positions (x, y, z) were calculated using swimming speed (v), the swimming depth (d) and the heading (θ) of recorded data using formulae below,

$$x_t = \sum_{n=1}^N \sqrt{v_t^2 - v_{z_t}^2} \cos \theta_t \quad (1)$$

$$y_t = \sum_{n=1}^N \sqrt{v_t^2 - v_{z_t}^2} \sin \theta_t \quad (2)$$

$$z_t = \sum_{n=1}^N (d_t - d_{t-1}) \quad (3)$$

Three-dimensional trajectories were reconstructed to summate of the 3D swimming vectors.

The component of tidal current was removed with high pass filter in the same manner of Komeyama et al. (2013).

3. Results and Discussion

The swimming trajectories with a color scale according to speed were reconstructed using measured

the swimming speed, the swimming depth and the heading (Figure 1). Bluefin tuna swam in alignment with the wall of the net cage in the daytime.

We compared to curvature of the swimming speed between day and night (Figure 2). We found that the outlier of swimming curvature at night. This imply that the fish exhibited sharply turn which imply the tuna did burst swimming at night in the cage. it might relate to collision of the reared fish with wall of cage. Therefore we should take measure to prevent from collision of the fish to improve the mortality of tuna aquaculture.

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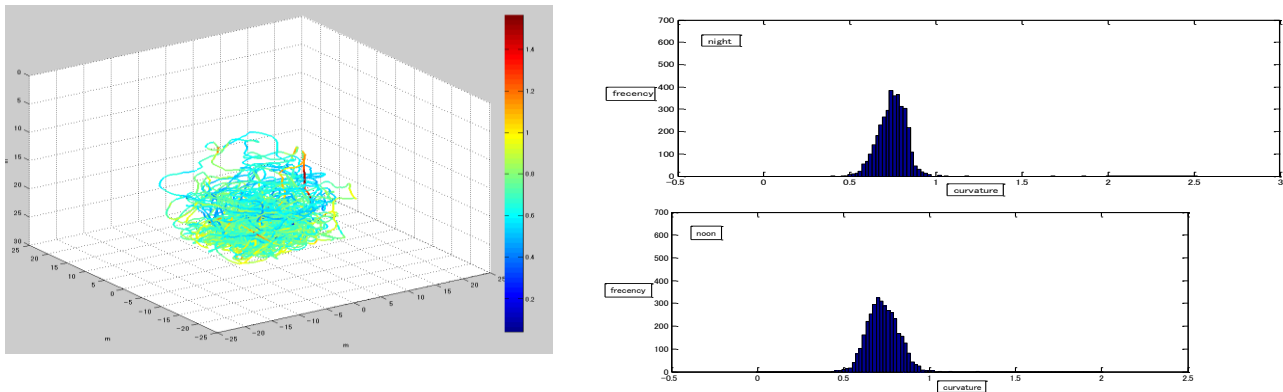


Figure 1 (Left) Typical result of Swimming trajectories

Figure 2 (Right) Histogram of curvature of the trajectories day and night

A comparative study on the development of giant clams *Tridacna noae* and *Tridacna maxima* (Bivalvia: Tridacnidae) in Taiwan

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1. Purpose

The aim of this study was to compare larval development of *T. maxima*, *T. noae* and hybrids of the two species.

2. Materials and methods

Mature giant clams collected from Dongsha were transported to the Marine Life Propagation Station in Penghu for experimental use. Eggs and sperms were sampled when giant clams spawned and artificial fertilization was conducted subsequently.

3. Results

The fertilized eggs of *T. maxima*, *T. noae* and hybrids, with diameters of $113.75 \pm 18.5 \mu\text{m}$, $99.04 \pm 8.1 \mu\text{m}$ and $116.3 \pm 6.2 \mu\text{m}$, were cultured in static water tanks at temperature of $27.8 \pm 0.3 \text{ }^\circ\text{C}$. Shell lengths of day-2 straight-hinge veliger of the 3 groups were $142.8 \pm 4 \mu\text{m}$, $147.8 \pm 5.7 \mu\text{m}$, and $151.9 \pm 6.7 \mu\text{m}$, respectively. At day-6, *T. maxima* and *T. noae* developed to D-veliger with shell lengths of $181.5 \pm 9.2 \mu\text{m}$ and $181.5 \pm 6.7 \mu\text{m}$. In contrast, the death of hybrids occurred at this stage. At 8 - 9 days after fertilization, shell lengths of *T. maxima* and *T. noae* reached to $194.5 \pm 4.3 \mu\text{m}$ and $193.6 \pm 5.0 \mu\text{m}$ and the settlement and metamorphosis started. At day-15, juveniles of *T. maxima* and *T. noae* had shell lengths of $198.1 \pm 14.7 \mu\text{m}$ and $200.2 \pm 9.4 \mu\text{m}$.

4. Discussion

Recent studies have shown that a cryptic species, i.e. *Tridacna noae* involved in *T. maxima* based on morphological and molecular evidences. The main difference is the pattern of mantle, i.e. radial for *T. noae* and spotted pattern for *T. maxima*. Besides, the numbers of eye spots on the edge of the mantle are also different. We tried to breed the hybrids of *T. maxima* and *T. noae* for the first time. Only a few eggs were fertilized successfully, but most of them died on the following day. Our cross-breeding experiment supports that *T. noae* is not a ecotype of *T. maxima* but a valid species.

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Reproduction and recruitment of land crabs (Decapoda: Brachyura) in Hsiangchiaowan, Hengchun Peninsula, Taiwan

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ABSTRACT

The purpose of the study is to investigate the reproductive population dynamics and megalopae recruitment of land crabs at Hsiangchiaowan coastal forest, Hengchun Peninsula, Taiwan. Larval release among female land crabs was regularly monitored within a 100 x 30-m rectangular portion of the supralittoral and littoral zones outside the coastal forest from late June to early September, 2010. Recruitment of megalopae of land crabs was also monitored within a surge channel from July 2010 to August 2011. A total of 4,072 larvae-releasing females belonging to 4 families and 13 species were collected during the study. The dominant larvae-releasing females were *Metasesarma aubryi*, *Gecarcoidea lalandii* and *Cardisoma rotundum*. The population of these dominant species was reduced significantly compared with the population size observed in 2002 and 2003. Moreover, the average carapace width of *G. lalandii* was smaller compared to the average size in 2003. Only megalopae of three land crab species, *M. aubryi*, *Cardisoma hirtipes* and *Scandarma lintou*, have been collected but without apparent recurrence of the rhythmic cycle. Our results showed that the breeding population of land crabs has decreased significantly in the past 7 years and the recruitment of megalopae is rare and unpredictable. Urgent conservation action is needed at Hsiangchiaowan coastal forest where is among the most important biodiversity hotspots for land crab fauna in Taiwan.

Key word : land crab, reproduction, recruitment, *Metasesarma aubryi*, *Gecarcoidea lalandii*, *Cardisoma rotundum*

Reproductive and migrations of the southern velvet shrimp *Metapenaeopsis Palmensis* in the waters off southwestern Taiwan

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ABSTRACT

The purpose of this study is to provide the reproductive biology information, describe the movement patterns and discuss possible environmental influences on the life histories of *Metapenaeopsis palmensis* in the waters off southwestern Taiwan via the investigation of carapace length frequencies and reproductive data. The study area covers the major fishing grounds for shrimp trawl fisheries, and it is located between the waters off Jiading southward to Fangliao, southwestern Taiwan. The reproductive season is from early spring to early summer with a single mode recruitment pattern in September in the study area. Newly recruited individuals were found in the deeper waters than adults. An inshore spawning migration for adults were also observed. Environmental factors such as temperature, salinity and precipitation, play an important role in affecting the life history of *M. palmensis* in different aspect. Water temperature raises in early spring trigger the gonadal development and the duration of the reproduction. Meanwhile, the combined effect of heavy precipitation, turbidity and salinity affect to decrease population size in rainy season. After the rainy season, the juvenile and subadults recruit to the coastal waters where become a stable environment with rich food supplied habitat for them to live.

Key words: spawning migration, life history, rainy season, recruitment, Kaoping River

Influences of light irradiation on production of fatty acids and carotenoids in thraustochytrids, genus *Aurantiochytrium*

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1. Purpose

Marine Eukaryotes, thraustochytrids have been explored because they can produce substantial biomass with rich docosahexaenoic acid (DHA) content and carotenoid pigments such as astaxanthin which is also known as functional food additives during short period. This study was undertaken to investigate the effect on production of fatty acids and carotenoids of thraustochytrids, genus *Aurantiochytrium*.

2. Materials and methods

Plate cultivation

Aurantiochytrium sp. strain ATCC 20888 and *A. limacinum* strains mh0186 and SR21 were grown on B1 agar plate medium at 28°C for 96 hours with or without fluorescence-light irradiation. The lawn of colonies on an agar plate was collected using a spatula to extract total lipids.

Growth test in Liquid medium

Strain ATCC 20888 showing significant colony pigmentation by light irradiation was subjected to further experiment. Strain ATCC 20888 was grown in a GY liquid medium with shaking (130 rpm) for 144 hours at 28°C with or without light irradiation. Every 24 hour, aliquot of cultures were collected to determine biomass and glucose concentration. After glucose in the cultures was exhausted, the cultured cells were collected to extract total lipid and total carotenoid.

Analytical procedure

The biomass was determined as dry cell weight by heating the wet cells at 105°C. The glucose concentration was determined using a commercial test kit (test wako C-II, Wako Pure Chemical Industries Ltd., Osaka, Japan).

The total lipid was extracted with chloroform-methanol solution (Folch et al., 1951). The fatty acids contained in the extracted total lipids were methyl-esterified with a methanol-HCl solution followed with heating at 80°C for 3 hours. The composition of methyl ester fatty acids (MEFAs) was analyzed by gas-chromatograph (GC-2014, Shimadzu co., Kyoto, Japan). The injection port and a flame-ionization detector were held at 250°C. The column temperature was initially held at 150°C and was programmed to be heated to 220°C at 2°C per min. Helium was used as the carrier gas.

Total carotenoid was extracted by ice-cold acetone in dark and determined according to the spectrophotometric method (Lichtenthaler, 1987). Total carotenoid was separated into each carotenoid by thin layer chromatography using a petroleum ether-acetone solution (7:3, v/v) as an eluent.

3. Results

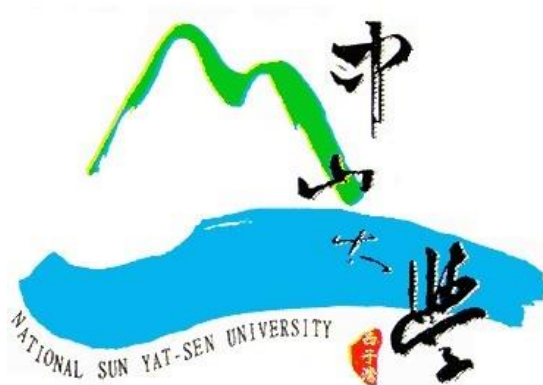
The pigmentation of strain ATCC 20888 on a plate was significantly enhanced by light irradiation, but no differences were observed in strains mh0186 and SR21. In the case of liquid culture, strain ATCC 20888 grew well by irradiation. The highest content of total fatty acid was 324.7 mg/g-DCW at 96 hours with light irradiation and 263.6 mg/g-DCW at 120 hours without light irradiation. Significant difference was observed between groups. Stearic acid and DHA were mainly detected as a saturated fatty acid and an unsaturated fatty acid, respectively (Fig. 1). In the case with light irradiation, the content of stearic acid (90.6 mg/g-DCW) and DHA (99.9 mg/g-DCW) was significantly higher than those without light irradiation (63.2 mg/g-DCW and 87.7 mg/g-DCW, respectively). The content of total carotenoids increased significantly by light irradiation (Fig. 2). In total carotenoids, only β -carotene was detected until 96 hours, and cantaxanthin and astaxanthin were also detected at 120 hours and 144 hours.

4. Discussion

This study represented that light irradiation improved the colony pigmentation of ATCC 20888 grown on an agar plate, but not in the case of strains mh0186 and SR21, which means the sensitivity to light is obviously different among strains of genus *Aurantiochytrium*. Furthermore, light irradiation enhanced the growth of strain ATCC 20888 and improved the production of total fatty acids and total carotenoids. Generally, pigments like carotenoids play a role to diffuse the damages of tissue and cells under the photo oxidative conditions. Thraustochytrids have large amount of unsaturated fatty acids (USAs) which are apt to become oxidative, and these pigments may have functions to protect the oxidation of USAs. However, there are no information about the relationship among light irradiation, growth of thraustochytrids and production of pigments and fatty acids except for this study. In further, it is needed to more clarify this point.

5. References

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