Faculty of Resource Science and Technology

Preliminary Evaluation on the Potential of Sea Urchins as Fish Attractant

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DECLARATION

I hereby proclaim that no portion of this thesis has been submitted in support of an application for another degree of qualification of this or any other university or institution of higher learning.

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Preliminary Evaluation on the Potential of Sea Urchins as Fish Attractant

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ABSTRACT

Five types of methods were prepared for this experiment. Each method contained gonad of *Diadema setosum* to use as natural stimulants for seabass. Five types of methods were raw gonad, gonad in powder form, gonad solution, raw wet agar strips coated with gonad in powder form and raw agar strips immersed in gonad solution. The experiment was conducted on seabass of 7.6 cm of total length by using one fish per tank. Feeding responses were classified into: (1) No reaction: fish remains on station without moving. (2) Orientation: rapid movement of the head pointing at food. (3) Approach: fish swims quickly towards food/odour. (4) Capture: fish nibble at food. (5) Ingestion: fish eats food. However, no feeding response was observed in control tank. For three types of method without raw agar strips, it showed that raw gonad method was the best respond time among the three methods and for another two methods with raw agar strips, it showed that raw wet agar strips coated with gonad in powder form was the best respond time. The experiment is useful for determining the suitability gonad of sea urchin to use as fish attractants. The study revealed that gonad of *Diadema setosum* can be used as fish attractants.

Key words: Sea Urchin, fish attractants, chemoreception, respond time, feeding response.

ABSTRAK

Lima jenis kaedah telah disediakan untuk eksperimen ini. Setiap kaedah mengandungi telur Diadema setosum *yang digunakan sebagai perangsang semulajadi untuk ikan siakap. Lima jenis kaedah tersebut adalah telur mentah, telur dalam bentuk serbuk, telur dalam bentuk cecair, jalur agar yang basah disalut dengan serbuk telur dan jalur agar yang direndam di dalam cecair telur. Eksperimen ini telah dijalankan ke atas ikan siakap berukuran 7.6 cm panjang dengan menggunakan satu ekor ikan untuk setiap tangki. Tindakbalas pemakanan telah diklasifikasikan kepada: (1) Tiada reaksi: ikan kekal tanpa bergerak. (2) Orientasi: pergerakan kepala yang pantas menghala ke arah makanan. (3) Pendekatan: ikan berenang cepat ke arah makanan/bau. (4) Tangkap: ikan menyentuh makanan. (5) Ditelan: ikan memakan makanan. Bagaimanapun, tiada pemerhatian dilakukan terhadap tangki kawalan. Untuk tiga jenis kaedah tanpa jalur agar, ia menunjukkan bahawa kaedah telur mentah mempunyai masa tindakbalas terbaik di antara tiga kaedah dan dua lagi kaedah dengan menggunakan jalur agar, ia menunjukkan bahawa jalur agar yang disalut serbuk telur mempunyai masa tindakbalas yang terbaik. Eksperimen ini berguna untuk menentukan kesesuaian telur landak laut untuk digunakan sebagai bahan penarik ikan. Kajian ini telah menunjukkan bahawa telur* Diadema setosum *sesuai untuk digunakan sebagai bahan penarik ikan.*

Kata Kunci: Landak laut, bahan penarik ikan, kemoresepsi, masa tindakbalas, tindakbalas pemakanan.

1.0 INTRODUCTION

Sea urchins can be found all over the oceans. There are 800 species of sea urchin inhabiting the ocean floor, scavenging for plants and animals (Pawson, 2007). In tropical regions, genera *Diadema* are the most abundant and ecologically important sea urchins (Carpenter, 1997). Its populations occur in all tropical seas to a depth up to 70 m (Mortensen, 1940 as cited by Lawrence, 2007). These sea urchins affect the ecological structure of coral reefs through bioerosion and substrata (Bak, 1990; 1994) and compete for space between corals and algae (Coyer *et al.*, 1993 as cited by Mapstone *et al.*, 2007). Furthermore, these sea urchins also are dangerous to divers because of their long spines are extremely sharp and painful. Besides that, *Diadema* spp. is eaten only by certain ethnic in Sabah. So, from letting the sea urchin populations to increase and causing damage to coral reef habitat, it is better to utilize these sources by using their gonads that is light yellow in colour as fish attractant.

There are many different types of fish attractants in the market today. Fish attractant is a product designed to attract and trigger the fish responses. It can be made from any types of materials. Generally, the base of attractant is liquid and oil. The attractant is dipped into or coated the bait. When the bait hook is placed in the water, the attractant from the bait spreads into the water. However, the standard attractants tend to float at the surface of the water and thus have limited effectiveness in accomplishing their intended purpose (Shumaker, 1991). Besides that, an advanced fish attractant is formed from the mixing of animal feed and flavouring that can attract the fish chemoreceptor organs.

Other than that, there are three different phases of food in food search behaviour that are initial period of excitement, wherein the fish is alerted to the presence of the stimulus. Secondly is exploratory phase to locate the source and lastly the consummation phase which the fish seeks to ingest the potential food (Wunder, 1927 as cited by Hara, 1992). While according to Atema (1971) as cited by Hara (1992), she further separates the final phase into food uptake and food ingestion.

Seabass is a fish that inhabiting the estuarine and fresh water zones of the coastal regions. It also can tolerates wide salinity changes and fetches higher market price in market due to its good taste. The qualities such as fast growth and good taste tempt aquaculturist to culture this fish. The euryhaline character makes it an ideal fish for coastal aquaculture since the fluctuation of salinity is very frequent especially in rainy season. This fish are carnivorous and prefer trash fishes (Singh *et al*., 2006).

Currently, peoples cannot see the usage of sea urchins. So, this study had verified that the products from gonads of sea urchins are suitable to be used as bait for fish attractant. Furthermore, this study also will help to score the responses of fish towards the food given. For this study, the samples of *Diadema* spp. were collected from Satang Island, the gonads were dissected immediately and brought to the laboratory to be freeze dried. Then the gonads were tested as fish attractant. The seabass fingerlings were used in this study to test whether they will attract to the products of the gonads or not.

From the preliminary observations towards trial experiments that had been carried out, the fishes showed positive responses. Different responses were shown by the fish as different amount of gonads were tested towards them. In addition, the success of this study also will help to improve the fishery sector.

Furthermore, this project is mainly to evaluate on the potential of sea urchins as fish attractant. The outcome of this study can be used in sport fishing enthusiast with the development of suitable fish attractant in the market. Therefore, the study was conducted in order to: 1) make fish attractant for artificial bait from the gonads of sea urchins and 2) obtain and record the responses of the fish towards the food given.

2.0 LITERATURE REVIEW

2.1 Fish Attractant

Fish attractant involves the techniques and equipment to persuade fish to bite on the artificial or natural bait. A wide variety of bait are used to stimulate the various senses of fish such as visual, electrical, pressure and others that replicate natural feeding patterns and stimulate aggressive attack responses. Fish attractant must be water soluble to be effective (Prochnow & Cihlar, 1998). Besides that, the commercial attractants have appeared in the last two decades with the increasing recognition by anglers of chemoreceptive powers of fish. The most common use of attractants by anglers is an external treatment for artificial lures. Three reasons commonly these treatments were used: (1) to chemically arouse and attract nearby fish, (2) to increase lure retention time once taken into the mouth, and (3) to mask potential feeding deterrents present on the lure (Hara, 1992).

Furthermore, the function of fish attractant is to enhance the effectiveness of the bait. It also must be cumbersome and messy to make fish aware and to lead them to the bait. A good fish attractant is it can make the attractant spreads in all directions from the bait when deployed in the water to make the fish around the area to be attracted to the bait (Shumaker, 1991). In addition, according to Tiwari and Singh (2007) as cited by Kumar and Singh (2010), the attractants also can be combined with the toxicants and has been observed that it is an effective tool to manage the pest.

Besides that, the functions of feeding stimulants are supposed to promote quicker food intake without disturbing the water quality and help the fishes at several feeding steps like initiation at longer distance ('attractant' effect), help the catching of the prey at shorter distances ('arrestant' effect), initiate the feeding via tasting ('incitant' effect) or promote continuation of the feeding ('stimulant' effect) (Mackie, 1982 as cited by Singh *et al.*, 2006). The free amino acids are reported to have attractant and arrestant effects as they diffuse in the water effectively and stimulate the olfactory bulb (Hara, 1973 as cited by Singh *et al.,* 2006).

2.2 Chemoreception of Fish

Chemoreception plays a major role in fishes that involves all aspects of life including feeding, prey detection, predator avoidance, species and sex recognition, sexual behaviour, parental behaviour and migration. Chemoreceptions are mediated by the ability to detect water-born chemicals and to react to these stimuli (Helfman *et. al.*, 2009).

In addition, chemoreception is needed by the fish larvae due to their limited vision (Blaxter and Stains, 1970) and the fact that chemical stimuli persist over large distance to serve as accurate indicators of food location (Sorensen and Caprio, 1997 as cited by Kolkovski *et al*., 2000). Chemoreception also restricted to those interactions involving primary or secondary chemosensory cells in the receiving organism (Mackie and Mitchell, 1981). Moreover, chemoreception can be divided into two basic types that are olfaction, (the sense of smell) and gustation, (the sense of taste).

Olfaction is a distance sense that enables the fish to locate and find the food or sexual partner or to avoid an enemy on a greater distance (Emde *et al.*, 2004). Other than that, olfaction is the results from the stimulation of sensory receptor cells in the olfactory organs which are innervated by the olfactory nerve (cranial nerve I). Figure 1 shows the lateral view of the brain and roots of the cranial nerves of a salmon (Helfman *et al.*, 2009).

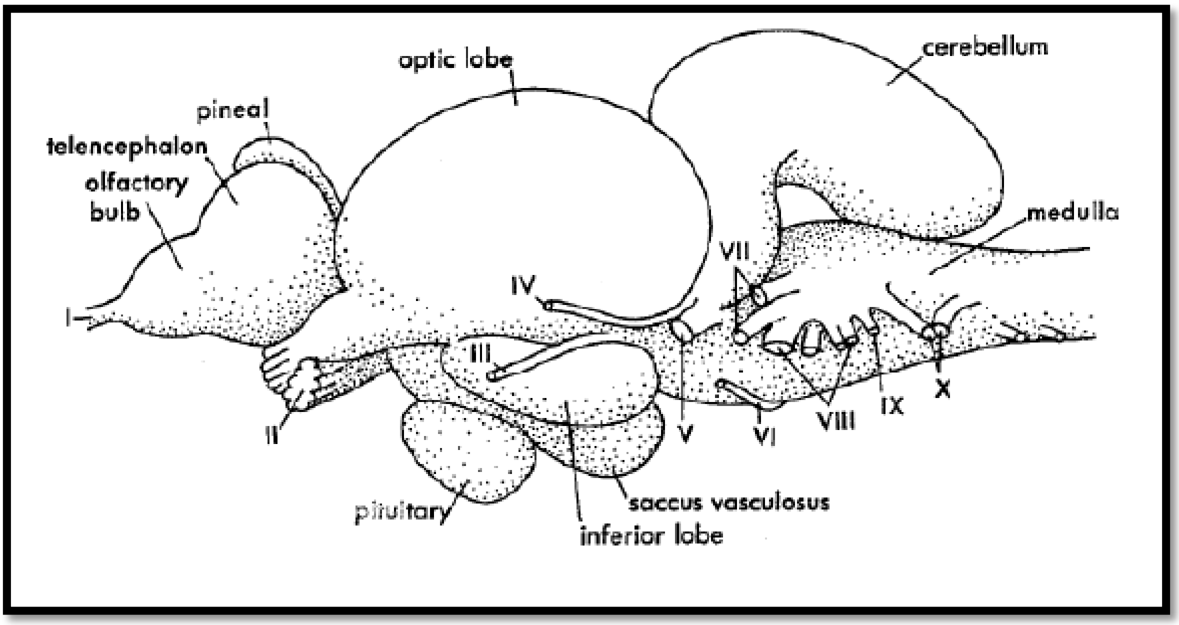

Figure 1: Lateral view of the brain and roots of the cranial nerves of a salmon: olfactory nerve (I), optic nerve (II), oculomotor nerve (III), trochlear nerve (IV), trigeminal nerve (V), abducens nerve (VI), facial nerve (VII), auditory nerve (VIII), glossopharyngeal nerve (IX) and vagus nerve (X).

The olfactory system of fish consists of the peripheral part of the olfactory organ proper including the olfactory nerve formed by the axons of the olfactory receptor neurons (ORNs) and the central part comprising the olfactory bulbs and the higher brain areas involved in processing of olfactory information. The peripheral olfactory system is able to detect thousands of smell stimuli that are utilized for food search, intraspecific and interspecific interaction, orientation and reproduction. Hence, the olfactory system plays an important role and if the system is damage it may decrease the health of fish and they cannot survive for a long time (Emde *et al*., 2004).

While gustation serves to detect by using taste and approve of the foodstuff that is near to the head or even taken up into the mouth (Emde *et al.*, 2004). It is mediated by taste buds and innervated by the facial, glossopharyngeal and vagus nerves (cranial nerves VII, IX, and X) as shown in Figure 1. Taste buds also are found in the mouth cavity, as well as in the gill cavity, on the gill arches and in some cases on the external surfaces of the body (Helfman *et al.*, 2009).

Besides that, there is another type of chemoreception that is common or general chemical sense. It is mediated by sensory receptors located on exposed body surfaces of a fish and better developed in scaleless, bottom living fish. Taste buds are not involved, instead the receptors are free nerve endings supplied by the spinal nerves. It is also low sensitivity compared to smell and taste (Helfman *et al.*, 2009).

2.3 Senses of Fish

Fish not only produce and respond to variety of noises, they also can sense the motions made by other fishes and in some cases the minute electric charges generated by the beating hearts of their prey. They can detect the amounts of various chemicals that tell them whether other fishes are nearby (Wilson and Wilson, 1992). Sensory systems also provide fishes with information from external environment, permitting appropriate responses to change the conditions. Furthermore, sensory cells act as signal transducers, receiving stimuli and convert to biological signals that transmitted via nervous system. The fish sensory systems include mechanoreception that consists of two components that are hearing and lateral line, visual chemoreception, electroreception and magnetic reception (Helfman *et al.*, 1997).

In the water, fish have different vision because the cornea of the fish is having the same refractive index as water. Their eyes have large lenses operated by powerful muscles that can bend the light rays in order to produce an image. Besides that, since the ocean absorbs daylight, the level of natural illumination declines rapidly with depth. To balance it, the fish eyes must be larger in order to gather in more light. The fish's eyes are also adapted to the colour of the light in the ocean. That colour changes with the variations in the depth of the water and with the amount and kind of organic matter floating in it. Blue light penetrates the deepest of water and things that observed at depth tend to appear blue in colour (Wilson and Wilson, 1992).

2.4 Feeding Behaviour of Fish

Feeding behaviour of fish is the interactions between fish and their environment, among each other and their own physiology. This feeding behaviour is influence by the environments such as temperature, light intensity, seasonality and activity cycles. Besides that, the feeding behaviour is actually acted as the indicator of appetite. Figure 2 shows how the process happens (Rumohr, 2011).

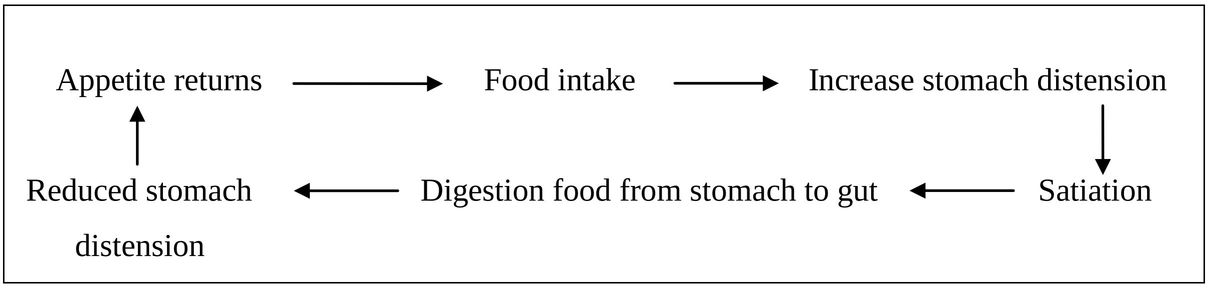

Figure 2: Indicator of appetite process.

Besides that, feeding behaviour can influence the growth, reproduction, health of fish and their response to the physiologic, environmental stressors and pathogens. This behaviour is also said to be a complex behaviour because it is closely related to the food intake. The food intake is one of the basic of fish metabolism. There are another two basics of fish metabolism that are digestion and transport of nutrients to various tissues. Several feeding behavioural responses are linked to many factors such as method of feeding, feeding habits, frequency of feeding, mechanism of food detection and lastly food preferences (Lall and Tibbetts, 2009). In addition, the feeding of fish should last within 5 to 10 minutes. It depend on how many fish in the tank and how fast the fish can capture and consume the food and the most important is what type of food is being feed (Axelrod and Burgess, 1979).

Furthermore, feeding behaviour composed of orienting responses, appetitive search swimming, reflex turning and consummatory behaviours such as reflex snapping or biting, oral manipulation, mastication and swallowing. Fish can detect their food by either visual or chemical senses in most cases. Visual stimuli enable the fish to swim directly at food items without the need of chemical stimulation, whereas olfactory that is taste stimuli will excite the fish to swim around to search the food (Valentincic and Caprio, 1994 as cited by Emde *et al.*, 2004).

The chemical that enable the fish to locate the food is amino acid. Amino acids are the building blocks of all organisms and present in the proteins. While free amino acids are dissolved in the cytoplasm. The balance between loss and uptake of amino acids from and into living organisms will determine the concentration of free dissolved amino acids in the water (Fergusson, 1980 as cited by Emde *et al.*, 2004). The concentrations of L-alanine and glycine in seawater are greater than 100×10^{-9}, whereas the concentrations of unstable amino acids in natural water such as L-cysteine may be as low as 1×10^{-9}. The fish enable to detect the presence of food when they detect the concentrations of amino acids is higher than the concentrations of natural background. Besides that, the chemoreceptors of fish also sensitive to aliphatic acids, nucleotides and bile salt (Marui and Caprio, 1992 as cited by Emde *et al.*, 2004). Those substances can also indicate food but the fish behaviour is not known.

2.5 Feeding Scores

There are many methods to scores the feeding responses of the fish. It is depends on what types of feed that were used. Stradmeyer (1989) used methods such as orientation, approach, capture, rejection and ingestion to score the feeding responses of the fish. While Singh *et al.* (2006) used methods such as no reaction, orientation, approach, capture – ingestion and capture – rejection to score the feeding responses of the fish. The feeding score methods such as changes in total swimming activity and the turning rates of fish head was used by Davis *et al*. (2006) and striking behaviour of fish method was used by Nakajima *et al.* (1989).

3.0 MATERIALS AND METHODS

3.1 Sources of Sea Urchins

About 40 individuals of sea urchin *Diadema setosum* were collected at Satang Island on 16 September 2011. The samples were collected by hand at subtidal area about five metre depth and were dissected immediately for the gonads. The gonads were packed together with seawater and brought back to the laboratory for further preparation and bioassay.

3.2 Laboratory Works

3.2.1 Storage of Samples

In the laboratory, the gonad samples were stored at -20^{o}C deep freezer. Later the gonads were thawed and sieved to reduce the excessive seawater inside the sample and stored again in a freezer with temperature -20^{o}C prior to freeze drying. Five small bottles of gonads were freeze dried using a freeze dryer machine (Labconco model 7755033 – 18L). Then, the dried gonads were grinded into powder form by using pestle and mortar.

3.2.2 Fish Attractants Preparation

There were five methods tested in this study and for each method the concentrations of gonad used were based on weight 0.1 g, 0.2 g, 0.3 g, 0.4 g and 0.5 g.

The gonad was weighed using an electronic balance (Shimadzu type, BL 220 H). The preparation methods of fish attractants were shown in Figure 3.

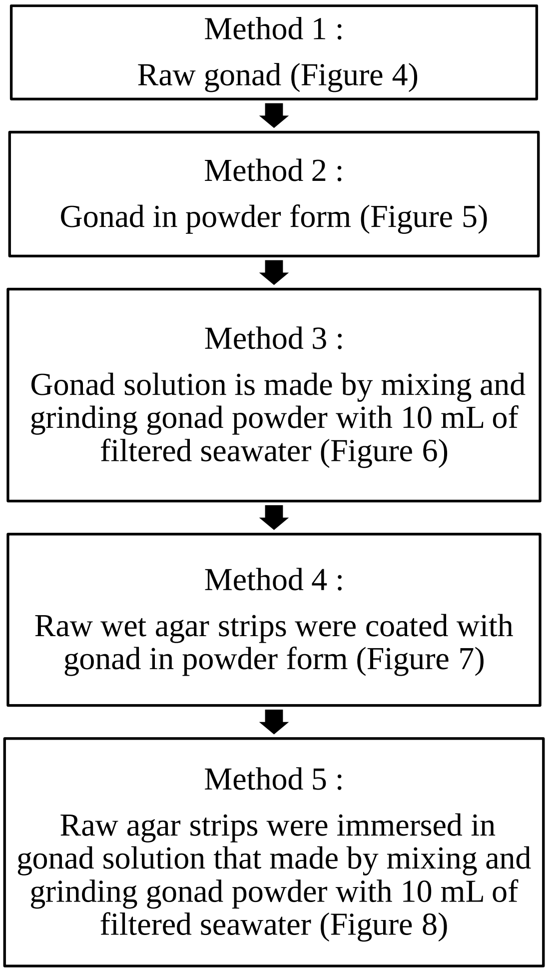

Figure 3: Preparation methods of fish attractants.

Figure 4: Raw gonad from concentration 0.1 g to 0.5 g (left to right).

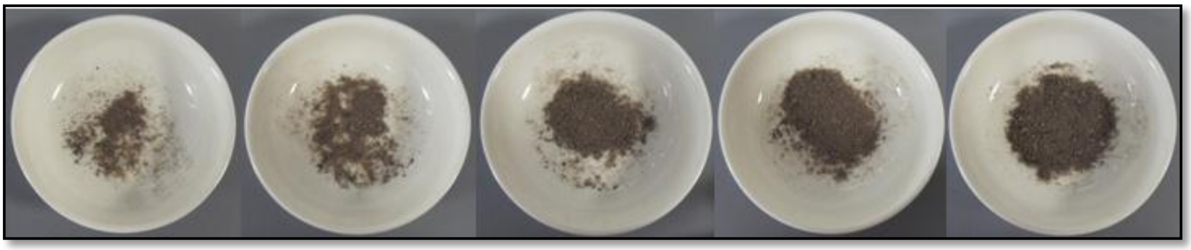

Figure 5: Gonad in powder form from concentration 0.1 g to 0.5 g (left to right).

Figure 6: Gonad solution from concentration 0.1 g to 0.5 g (left to right).

Figure 7: Raw wet agar coated with gonad in powder form from concentration 0 g to 0.5 g (left to right).

Figure 8: Raw agar strips immersed in gonad solution from concentration 0 g to 0.5 g (left to right).

3.2.3 Bioassay

A total of 30 seabass fingerlings of 7.6 cm total length (Figure 9) and seawater were obtained from Santubong Fisheries Department. They were held for acclimatisation in rectangular glass tank (47 × 45.3 × 59.5 cm) containing seawater (24 – 25^{o}C, 20 – 21 PSU) for one week in the Aquatic Invertebrate Laboratory. The tank was provided with continuous aeration and water exchange was done every two to three days. During the acclimatisation period, the seabass were fed with pellets twice a day. All bioassays were conducted by using one fingerling per tank.

Figure 9: Seabass fingerling (Total length = 7.6 cm).

Three rectangular glass tanks (41 × 30 × 31 cm) with the capacity of 29 litres were used for the bioassays. The set up of the tanks as shown in Figure 10. As for trial, small pieces of gonads in the form of powder were introduced into the tank to observe the response of fish. At the start of the bioassay, three fishes which had been acclimatised were stocked in bioassay tanks and later subjected to bioassay. The fishes were fed once daily with pellets between 1600 to 1700 hours. The bioassays were performed between 1400 to 1600 hours.

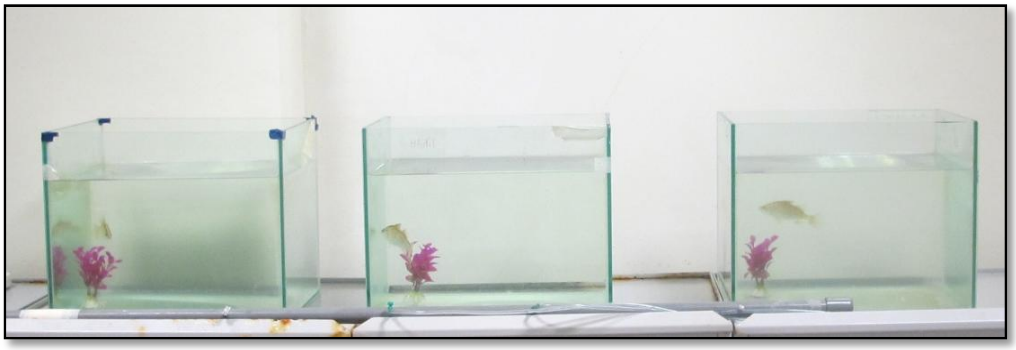

Figure 10: The set up of the tanks for bioassay.

Each method took three days to complete the bioassay. Each fish were tested three times for each concentration. For method 1, 2 and 3, the three tanks resulted 45 observations of responses per method and for method 4 and 5, the three tanks resulted in 54 observations of responses per method. In each tank of all methods tested, one artificial plant was put as the initial point of observations (Figure 11). The water in bioassay tanks was exchanged after the bioassay in one day completed. All the observations were recorded using a digital camera (Canon, PowerShot G12).

Figure 11: Artificial plant in the tank as the initial point of observation.

For method 1, the different concentrations of raw gonads were introduced in bioassay tanks. For method 2, the gonads in powder form were scattered on the surface of the tank. In method 3, three droplets of attractant were introduced into each of the tank. Raw wet agar strips that coated with gonad in powder form for method 4 and raw agar strips that immersed in gonad solution for method 5 were introduced to the fish. All of the attractants tested were introduced at the right corner of the tank.

The feeding responses will be scored from one to five adapting methodology of Stradmeyer, 1989 and Singh *et al.*, 2006 and expressed as following: (1) No reaction: fish remains on station without moving. (2) Orientation: rapid movement of the head pointing at food. (3) Approach: fish swims quickly towards food/odour. (4) Capture: fish nibble at food. (5) Ingestion: fish eats food. The first reaction of respond time of fish were observed and recorded.

3.3 Environmental Parameters

The temperature of water in the tanks was measured using water proof thermometer (Traceable model 4039) and the salinity of water was measured using salinometer (Milwaukee, MA887).

4.0 RESULTS

4.1 Respond Time of Fish

The bioassay using seabass fingerlings were tested against five different types of method. The objective of the experiment is to find the fastest respond time of fish towards the attractants given. The concentration that showed the fastest respond time was the best time to prove that gonad of sea urchin has the potential to be used as fish attractant. Besides that, every fish showed different response based on the observation in the experiment.

4.1.1 Respond Time of Fish Towards Raw Gonad

In the first experiment, different concentrations of raw gonads were used to be tested with the fish. The first reaction showed by three different fish were presented in Figure 12. For each concentration, a fish was tested only once a day and each fish was repeated with three trials of the same concentration on three consecutive days.

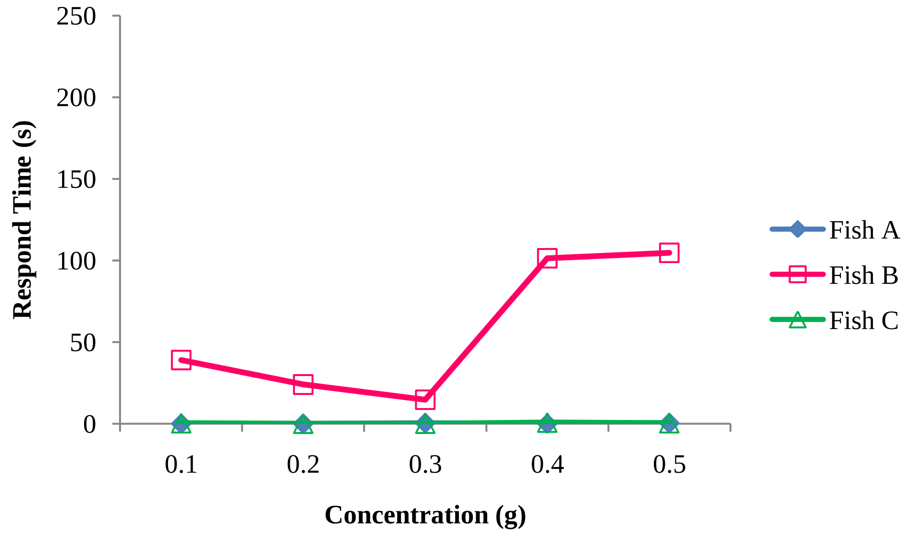

Figure 12: Respond time of the first reaction showed by three different fish towards the raw gonad.