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QUALITY ASSESSMENT OF GIANT FRESHWATER PRAWN (*Macrobrachium rosenbergii*) IN ICE STORAGE CONDITION COLLECTED FROM SELECTED FARMS AND DEPOTS

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ABSTRACT

This study was conducted to evaluate the quality changes in an important crustacean species giant freshwater prawn, *Macrobrachium rosenbergii* (locally called golda) at various stages of handling. The samples under study were obtained from farms and depots of four golda producing districts viz. Khulna, Bagerhat, Jessore and Norial area by determining organoleptic and biochemical aspects. The golda samples stored in ice immediately after harvest were in acceptable condition for seven days after harvesting period while the samples obtained from depots from all stations were in acceptable condition for 4 days only. There was a large fall of protein solubility and ATPase activity in all the samples obtained from farms and depots. The fall of solubility and ATPase activity was faster in samples obtained from depots compared to the samples obtained from farm immediately after harvest. The large fall of protein solubility and ATPase activity of samples indicated the denaturation of protein during ice storage.

Key Words: Quality assessment, *Macrobrachium rosenbergii,* Ice storage, Protein solubility, ATPase activity

INTRODUCTION

Freshwater prawn, golda (*Macrobrachium rosenbergii*) is the largest one among the freshwater prawns and is preferred by the consumers at home and abroad for its taste, size and color. Bangladesh is a major exporter of *M. rosenbergii* caught from natural sources but with the increasing demand in the international market, golda farming has been expanding rapidly throughout the country. Over the recent past, there has been a notable increase in the export prices of the Bangladeshi prawn particularly in USA, Japan, and European markets (Ahmed, 2001).

Shellfish muscle is known to contain higher percentage of sarcoplasmic and myofibrillar protein and lower amount of stroma protein, higher percentage of unsaturated fatty acids that are degraded quickly through autolytic, microbial and oxidative spoilage.

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Assessing and selecting for quality of prawn is of great importance to produce a level of quality which will satisfy both the customer and statutory food legislation. Over the years, many different methods of quality assessment have been developed and investigated in an attempt to determine the most suitable index for use in quality control testing. It has been stated frequently that no single method is generally reliable for assessment of freshness and spoilage in seafood. Nevertheless, numerous microbiological, chemical, biochemical, as well as other instrumental methods, are appropriate for this purpose as long as their range of applicability in terms of raw material, preserving parameters, and storage conditions are realized and respected. The objectives of this study were to evaluate the quality changes of *M. resenbergii* collected from different stations through sensory and biochemical techniques.

MATERIALS AND METHODS

M. rosenbergii samples were collected from selected farms and depots of Dumuria of Khulna, Bagerhat Sadar of Bagerhat, Avoynagar of Jessore and Kalia of Norail. The samples were transported to the laboratory of Department of Fisheries Technology, Bangladesh Agricultural University, Mymensingh in iced condition in an insulated box to assess the degree of freshness by organoleptic and biochemical methods.

Organoleptic assessment

For organoleptic quality assessment of Golda on the basis of odor, texture, colour (with shell), colour of flesh and general appearance of prawn, a five member panel was constituted. The panel members included Faculty members of the Faculty of Fisheries, Bangladesh Agricultural University, Mymensingh. Qualities of the collected prawns were assessed by using the descriptive scheme provided by Howgate *et al.* (1992). The guidelines used to assess the quality are given in Table 1.

Each attribute is scored from 1 to 5 (typically) by novice or experienced assessors with high score 25 indicating the best quality (excellent), 24-22 = very good, 21-19 = good, 18-14 = acceptable, 13-08 = bad and 7-5 = very bad. The sum of all attribute scores is called demerit points, or grade points, and this value decrease linearly with storage time in ice. The direct relationship between grade point scores and storage time makes it easy to calculate remaining shelf life of fresh prawn when stored at 0°C (in ice).

Biochemical Assessment

Measurement of pH

Two grams of peeled prawn sample was homogenized with 10 ml distilled water in a blender and the pH was measured using a pH meter (Corning Model 250) for 30-60 seconds**.**

Total volatile base nitrogen (TVB-N) determination

Total volatile basic nitrogen (TVB-N) of the samples was determined according to the method described by the Official Journal of the European Communities (EC, 1995). Briefly,

the procedure is as follows: exactly 10 gm of the ground prawn sample were weighed in a suitable container and mixed with 90 ml of 6% perchloric acid. The samples were homogenized for two minutes with a blender. This was done in cool condition (2-6°C).

Hundred ml of the extract was put in Kjeldahl flask and 20 ml of 20% sodium hydroxide (NaOH) solution and few glass beads were added in the flask. The distillate was fixed up in a conical flask with 50 ml 3% boric acid solution containing 1-2 drops of mixed indicator. After boiling the flask for 15-20 minutes the distillate accumulated in conical flask which was titrated with 0.01N hydrochloric acid (HCl). The TVB-N is calculated by the following formula:

$$\text{TVB-N (mg/100g samples)} = \frac{\text{ml. of titrant} \times 0.14 \times 2 \times 100}{\text{Sample weight}}$$

Table 1. Determination of the grade points or defect points

Characteristics of whole prawn	Defect characteristics	Defect points
A. Odour	Natural odour	5
	Neutral odour	4
	Slight sour odour	3
	Ammonical odour	2
	Rotten odour	1
B. Colour (with shellon)	Natural colour	5
	Slight discolour/ slight pinkish	4
	Brownish red	3
	Discolour	1
C. Colour of flesh	White colour of fresh prawn	5
	Slight pink colour	4
	Pink colour	3
	Dull/discolour	1
D. Texture	Firm, consistent and elastic	5
	Moderately soft and some loss of elasticity	4
	Some softening	3
	Soft and Watery	2
	Flesh with juice	1
E. General appearance	Bright shining and iridescent	5
	Slight dullness and loss of brightness	4
	Definite dullness and loss of brightness	2
	Dull	1

Determination of solubility and ATPase activity from myofibrils

Myofibrils were prepared from muscles immediately after excision according to Perry and Grey (1956) with slight modification. The muscle was chopped by a meat grinder and chilled minced muscle (50g) was homogenized for 1 min in 5 volumes of 39 mM borate

buffer (pH 7.1) containing 25 mM KCl and 0.1mM DTT. The homogenate was centrifuged for 15 min at 600 x g. The residue obtained was again homogenized and centrifuged for 15 min. The light-colored upper layer of the residue consists of myofibril was recovered with small volume of 39mM borate buffer (pH 7.1) containing 0.1M KCl and 0.1mM DTT. The suspension was centrifuged for 15 min to remove the supernatant. Myofibrils were diluted with 4 volumes of 39 mM borate buffer (pH 7.1) containing 0.1M KCl and 0.1mM DTT, and coarse materials were removed by centrifuging at 400 × g. The suspension was centrifuged again for 15 mins. at 600 × g to precipitate the myofibril myofibril. After washing the pellet three times in the same way, myofibrils were suspended with a desired volume of 39 mM borate buffer (pH 7.1) containing 0.1M KCl to make a concentration of 10-15 mg/ml. Then myofibrillar proteins were extracted from isolated myofibrils with 0.6M KCl-0.03 M Tris-HCl at pH 7.5. The suspension was stirred gently and kept over night at 4^{o}C. Then the solution was centrifuged at 900 × g for 30 min and protein content in the supernatant was determined by the Biuret method (Gornall *et al*., 1949).

Assay of specific ATPase activity

In this study Ca^{2+}- ATPase assay was determined. The reaction mixture for the Ca^{2+}-ATPase assay contained 25mM Tris, 5mM $CaCl_2$, 0.1M or 0.5M KCl and 0.25 mg myofibril per ml. After preparation of the reaction mixture, an appropriate quantity of myofibril suspension was pipetted to the reaction mixture followed by 2 mins. pre-incubation. The reaction was started by the addition of 1mM ATP and then 2 ml portion of the reaction mixture was withdrawn at different time intervals. To stop the reaction 1ml of 15% tri-chloro-acetic acid was added. The supernatant obtained by 5min centrifugation at 3000 × g was analyzed for the liberation of inorganic phosphate (Pi) by a method described by Fiske and Subba Row (1925).

RESULTS AND DISCUSSION

Organoleptic Assessment

The organoleptic characteristics are judged by 5 member panel test. The prawns those were collected from the farms were analyzed and assessed every alternate days and those from depots assessed every days to determine the grade points and quality status.

Total grade points to the samples those were stored in ice at farm level immediately after harvesting was 25, 21, 17, 14 and 11 on 1st, 3rd, 5th, 7th and 9th day respectively. Sample was excellent in quality, at the 3rd day the samples was good and on 5th and 7th days the quality deteriorate some what but was in acceptable condition. On the 9th day the quality was found completely deteriorated and crossed the point of rejection. Thus, the samples collected from the farm were in acceptable condition up to seven days. The samples obtained from depots were found in acceptable condition for four days only. Total grade points on 1st, 2nd, 3rd, 4th and 5th day was 23, 19, 16, 14 and nine respectively. At the 5th day of observation the samples were unacceptable as a result of spoilage. From this assessment it is obvious that the quality losses of prawns occurred at various stages due to rough handling and delay in icing after harvest.

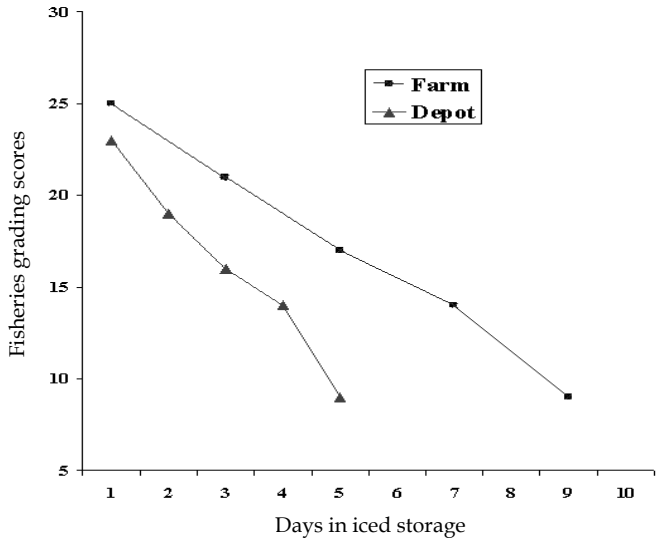

Fig. 1. Mean differences of the degree of Freshness of *M. rosenbergii* collected from different farms and depots

The pattern of organoleptic quality changes of prawn of farms can be roughly divided in to four phases corresponding to the periods of 0 to 1, 2 to 4, 5 to 7 and 8 to 9 days in ice. In phase I, the prawns were in very good condition with fresh bright shining and iridescent appearance, firm consistent and elastic texture, characteristics of white colour and natural shell odour and colour. In phase II, there was slight dullness and loss of brightness, slightly softening and some loss of texture, color of flesh and shell turned into slight pink and neutral odour. In phase III, there was considerable loss of brightness and softening of texture. The flesh was neutral but had no off odour. In phase IV, there was sign of early spoilage with sour odour and the shell became discolored. In the latter part of this phase, the prawn began to taste stale and its appearance began to show obvious signs of spoilage and had unpleasant smell.

Literatures are available about the oraganoleptic quality assessment suggestingat the shelf-life of prawn during ice storage which from species to species, due to factors like chemical composition, ambient temperature in which they are kept, post harvest handling and icing (Barlie *et al.,* 1985; Reilly *et al*., 1985; Dawood *et al*., 1986; Shamshad *et al*., 1990; Fonseka and Ranjini, 1994; Kodoria and Rojas, 1996; Rahman *et al.,* 2001a and Rahman *et al.,* 2001b). Shelf-life of shrimp ranged from 7 hours at 35^{o}C to 13 days at 0^{o}C (Shamshad *et al*., 1990). At ambient temperature shrimps were rejected after 12 hours while the shrimp held in ice were rejected after 15 days storage (Fonseka and Ranjini, 1994). In the present study sensory evaluation indicated that raw prawns of the farm had excellent freshness during the first three days of ice storage and was in acceptable condition for up to seven days which is more or less similar to the results obtained by Rahman *et al.*, (2001b). They reported that prawn those kept in ice immediately after harvesting remain in acceptable

condition for seven days. In another study they (Rahman, *et al.*, 2001a) determined the quality changes of prawn in ice and found that head-on prawns remain in acceptable condition in terms of commercial standard for up to 6 days, while headless ones for seven days.

The samples obtained from the depot were found acceptable for up to 4 days only. This shortened shelf-life may due to improper and delay icing of the prawn before transporting to the depots. Because Rahman *et al.* (2001b) found that the farms do not have sufficient facilities of immediate icing of the catch and therefore remain at ambient temperature for 4 to 12 hours prior to icing whereas delaying of 4, 8, and 12 hours in icing shortened the shelf life to 3, 2, and 1 days, respectively.

Biochemical Assessment

pH of the ice stored samples

Studies were conducted with *M. rosenbergii* obtained from culture farm and depot. The initial pH value of prawn sample from farm and depot were 6.8 and 6.5 respectively which gradually increased with the lapse of storage period. The pH value of the samples obtained from farm reached slightly beyond acceptable level after 6 days of storage. On the other hand, the samples obtained from depot were acceptable for up to 4 days in ice and during this period the pH value reached to 7.38 which are beyond the acceptable limit of 7.25. The decline in pH in early post-mortem muscle is due to the gradual hydrolysis during the first few hours of glycogen to lactic acid. The decline in pH is also accompanied by the natural post-mortem stiffening called rigor-mortis (Fig. 2).

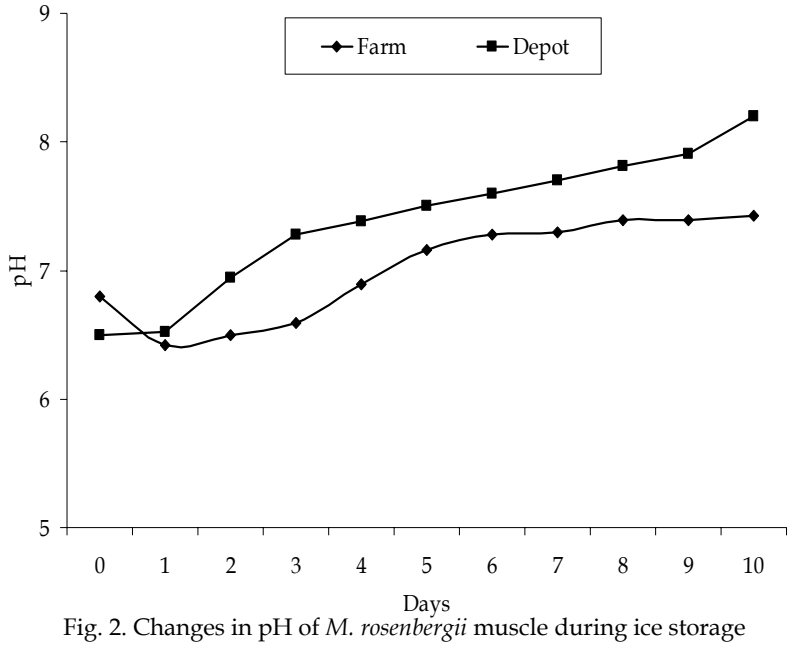

Fig. 2. Changes in pH of *M. rosenbergii* muscle during ice storage

Available reports suggested that increase in pH value of the collected samples during ice storage is due to accumulation of some basic nitrogenous substances like TMA and ammonia by the action of enzymes and microorganisms. Rahman *et al*. (2001b) reported that the pH of prawn over the period increase gradually from 6.36 immediately after catch to 8.0 after 10 days of storage in ice. This indicated a similarity to the result of the present study.

TVB-N of the ice stored samples

The mean initial TVB-N values of the samples collected from farms and depots were 5.8 and 6.2 mg/100g respectively, which also continuously increased during the progress of storage. In the samples collected from the farm, TVB-N value reached upper limit of 25 mg/100g after 6 days of storage while in samples collected from depot, TVB-N value reached beyond upper limit after 4 days of storage. There was a close relationship between TVB-N values and pH values where both the values increased with storage period. Another important relationship was between TVB-N values and organoleptic assessment of Golda during ice storage. Total freshness score or demerit score of the organoleptic assessment inversely changed with the TVB-N values of the collected samples (Fig. 3).

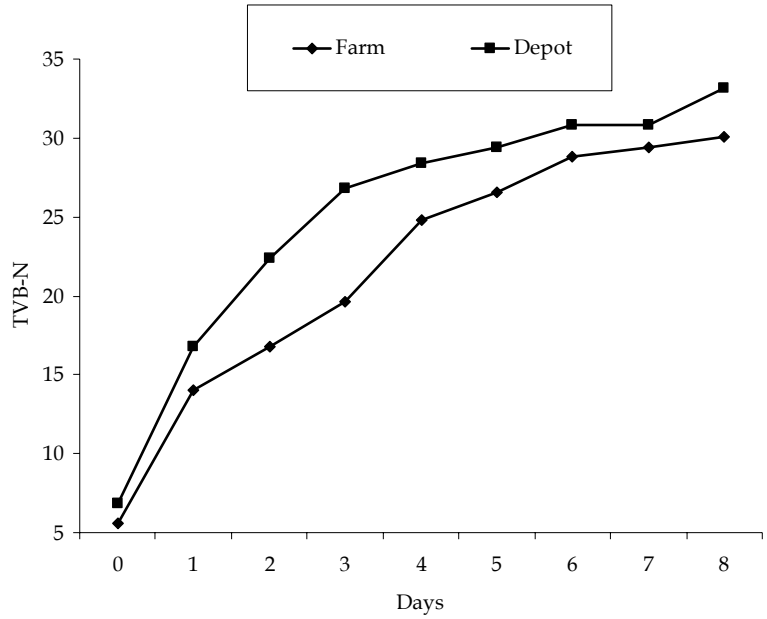

Fig. 3. Changes in TVB-N (mg/100g) values of *M. rosenbergii* muscle during ice storage

The available reports suggested that, the upper limit of 30 mg TVB-N/100g is considered as acceptable for finfish such as cod, haddock, eel and sea pike. The values fluctuated during ambient temperature and subsequent storage in ice. The fluctuation levels of TVB-N are probably due to the washing effects of ice during storage (Cobb *et al.*, 1976). The TVB-N values of samples stored at 0°C were lower than the recommended limits (30 mg

N/100g), even after 10 days storage. However, in samples stored at 5°C, this value found after 5 days (Leitao and Daniel, 2000).

The TVB-N values increased considerably with the storage period, and after 7 days, the TVB-N values were found within the recommended limit of 30 mg/100g in iced stored prawn (Rahman *et al.,* 2001b). This result revealed similarity to the result of the present study with the prawn samples collected from different farms of the country.

Changes in the myofibrillar protein solubility during ice storage:

The mean solubility and ATPase activities of the collected samples from different stations were also assessed to determine the degree of denaturation or degradation of the myofibrillar protein in ice storage condition. Studies conducted with the ice-stored *M. rosenbergii* showed that the mean initial myofibrillar protein solubility of the farm samples was 87%, which decreased to 53% during 10 days of ice storage. Similarly initial myofibrillar protein solubility of samples obtained from depot was 77% which also declined to 43% after 10 days of storage. The present study indicated that the protein solubility falls with the length of the storage period in ice storage condition. Moreover, the mean solubility was found significantly lower to the samples collected from depot rather than those from four farms. The decrease in the solubility of the protein may be the result of the proteolytic enzymes activity on the muscle protein (Fig. 4).

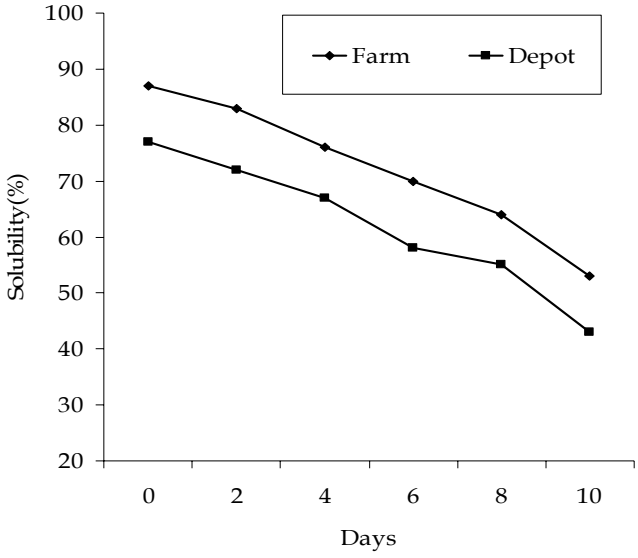

Fig. 4. Changes of Myofibrillar protein solubility of *M. rosenbergii* during ice storage

ATPase activity during ice storage

ATPase activity of the myofibrillar protein was determined in presence of 0.1M KCl with the prawn samples collected from different samples of farms and depots. The mean Ca^{+2}-ATPase activity of the myofibrillar protein was found to 0.482 µmol-pi/min. mg in the

prawn collected from farms, which was found to decline sharply to 0.287 µmol-pi/min. mg at the end of the 10 days of iced storage condition. Initially the mean ATPase activity of the myofibrillar protein of the samples from depot was found more or less similar to the value obtained in the farm sample which was 0.426 µmol-pi/min. mg but it was found to decline rapidly during ice storage and at the end of the 8 days of ice storage condition the result was 0.254 µmol-pi/min./mg. Thus, the rapid loss of Ca^{+2}ATPase activity was more in the samples collected from depot than those from farm. Reduction of the ATPase activity of the myofibrillar protein during ice storage occurs as a result of denaturation of protein (Fig. 5).

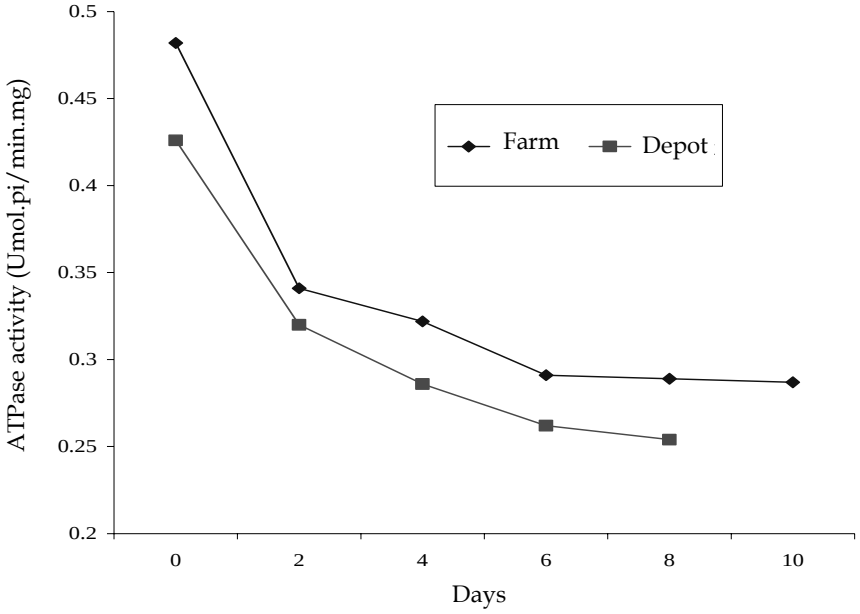

Fig. 5. Changes in mean ATPase activity of Myofibrollar Protein of *M. rosenbergii* obtained from samples of farms and depots

The results obtained from the present study are in agreement with those reported for various fish and shellfish species (Kamal *et al.*, 1990; Yasmin *et al.,* 2000; Rahman *et al*., 2001c). Yasmin *et al*. (2000) reported that myofibillar Ca^{2+}-ATPase activities of catla in presence of 0.1 M KCl were remained unchanged upto the first 5 days of storage and then gradually decreased along with the storage period and at the end of the 20 days of storage the activity declined to about 40% of its initial level. On the other hand, there was little or no change of $Ca2^{+}$-ATPase activities of myofibrillar protein in the presence of 0.5 M KCl throughout the storage period. In a similar study, they also reported that in case of mrigal, the initial Ca^{2+}-ATPase activities in the presence of 0.1 M KCl and 0.5 M KCl were 0.44 and 0.11 µmol-pi/min. mg respectively, which declined rapidly during 10 days of storage and after 20 days of storage, the activity decreased to 60 and 20% respectively.

Rahman *et al.* (2001c) studied that the initial Ca^{2+}- ATPase activities of *M. rosenbergii* in presence of 0.1 M KCl was 0.46 µmol-pi/min. mg which slightly decreased to 0.35 µmol-pi/min. mg after 10 days of storage, while there was little or no change in Ca^{2+}- ATPase activities in the presence of 0.5 M KCl. On the other hand, the mean ATPase activity of the prawn obtained from different samples in the present study from farm was 0.482 µmol-pi/min. mg and from depots 0.426 µmol-pi/min. mg, which indicated no significant differences to the reported results.

Determination of quality assessment of *M. rosenbergii* by organoleptic and biochemical methods indicate that its shelf-life was higher in samples obtained from the farms than depots. This is an indication that considerable loss of quality during different stages of handling from farm to depot level. The government has already taken initiative through Department of Fisheries (DoF) to reduce such quality loss, which will be helpful to reduce such loss and simultaneously increase the market price beneficial for its producers.

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