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# QUALITY AND SHELF LIFE OF THE ADDUCTOR MUSCLE OF LION'S PAW SCALLOP *Nodipecten subnodosus* TRANSPORTED AND STORED WHOLE IN REFRIGERATION

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## SUMMARY

The effect of refrigerated transport during 48h and four days storage on the quality and shelf life of the adductor muscle of whole lion's paw scallops (*Nodipecten subnodosus*) was evaluated. Proximal composition, ATP and related products, K-value, total volatile bases (TVB-N) and trimethylamine (TMA-N), pH, and microbiological analyses were quantified. The muscle was found to be lean and high in protein. Levels of muscular ATP were initially low and the K-value increased linearly, but accord-

ing to this parameter the product was considered edible until the end of the storage period. With respect to TVB-N and TMA-N, the allowed limits were not exceeded. The pH level showed no significant variations during storage. According to TVB-N, TMA-N values and microbiological analyses performed, adductor muscle proved to be innocuous after four days under the conditions of transportation and storage utilized.

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## Introduction

The lion's paw scallop *Nodipecten subnodosus* constitutes one of the most important fishery resources on Mexico's Baja California Peninsula. The commercial importance of this scallop rests on the size of the organisms (22cm tall) and their weight (250g at 5 years' growth), the price (USD16/kg in the international market), flavor, and the production of their adductor muscle. Its production is concentrated in the Ojo de Liebre Lagoon, state of Baja California Sur (BCS), and in recent years it has stabilized at 100ton/year. Another important characteristic of this

species is its high growth rate, as it attains a size of 7cm (commercial size) in only eight months. For the aforementioned reasons, fish farmers have become interested in producing this species and significant efforts are underway to consolidate its cultivation (Massó-Rojas *et al.*, 2001; Pacheco-Aguilar *et al.*, 2001; Maldonado-Amparo *et al.*, 2004).

While in both Mexico and the USA the principal product consumed is the fresh refrigerated adductor muscle, other countries, mainly in Asia and Europe, have a high demand for the whole shelled bivalves. These products are usually consumed after being cooked

in their shells, but are also served raw in dishes of the *sushi* or *sashimi* type (Paust and Rice, 2001; Pacheco-Aguilar *et al.*, 2001).

In terms of the nutritional value of pectinids, the main edible portion is the adductor muscle, which has a high protein content and a high biological value, comparable to that of casein and meat. Moreover, it is classified as lean food due to the fact that its lipid content is  $\leq 0.7\%$  (Ocaño-Higuera *et al.*, 2001).

In addition to the nutritional aspects, another factor that must be considered in the seafood product market so as to determine the optimum means of distributing this

scallop, maximize its value, and promote its consumption, is the quality and innocuous nature of the final product. Like many other aquatic organisms, upon their death the pectinids undergo a series of biochemical changes that strongly impact the quality and shelf life of finished products. These postmortem changes can be studied by monitoring certain parameters and indicators of quality and freshness (Sikorski *et al.*, 1990; Abbas *et al.*, 2008; Ocaño-Higuera *et al.*, 2009; 2011; Liu *et al.*, 2010).

At present there is no feasible and established strategy for exporting and distributing this particular pectinid in its

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## KEYWORDS / Lion's Paw Scallops / Muscle / Quality / Shelf Life /

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## CALIDAD Y VIDA DE ANAQUEL DEL MÚSCULO ABDUCTOR DEL PECTÍNIDO *Nodipecten subnodosus* TRANSPORTADO Y ALMACENADO ENTERO BAJO REFRIGERACIÓN

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### RESUMEN

Se evaluó el efecto del transporte bajo refrigeración durante 48h y almacenaje por cuatro días sobre la calidad y vida de anaquel del pectínido mano de león (*Nodipecten subnodosus*). Se cuantificó la composición proximal, el ATP y compuestos relacionados, el valor K, bases volátiles totales (BVT-N) y trimetilamina (TMA-N) y el pH, y se realizaron análisis microbiológicos. Se encontró que el músculo era magro y de alto contenido proteico. Los niveles de ATP fueron inicialmente bajos y el valor K aumen-

tó linealmente, aunque de acuerdo a este parámetro el producto se consideró comestible hasta el final del periodo de almacenamiento. Los límites permitidos de BVT-N y TMA-N no fueron excedidos. El pH no mostró variaciones significativas durante el almacenamiento. De acuerdo a los valores de BVT-N y TMA-N obtenidos y a los resultados de los análisis microbiológicos, el músculo abductor probó ser inocuo tras cuatro días bajo las condiciones de transporte y almacenaje utilizadas.

## QUALIDADE E VIDA EM PRATELEIRA DO MÚSCULO ADUTOR DO PECTÍNIDO *Nodipecten subnodosus* TRANSPORTADO E ARMAZENADO INTEIRO SOB-REFRIGERAÇÃO

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### RESUMO

Avaliou-se o efeito do transporte sob-refrigeração durante 48h e armazenagem por quatro dias sobre a qualidade e vida em prateleira da vieira pata-de-leão (*Nodipecten subnodosus*). Quantificou-se a composição proximal, o ATP e compostos relacionados, o valor K, bases voláteis totais (BVT-N) e trimetilamina (TMA-N) e o pH, e se realizaram análises microbiológicas. Encontrou-se que o músculo era magro e de alto conteúdo proteico. Os níveis de ATP foram inicialmente baixos e o valor K

aumentou linearmente, mesmo que de acordo a este parâmetro o produto se considerou comestível até o final do período de armazenamento. Os limites permitidos de BVT-N e TMA-N não foram excedidos. O pH não mostrou variações significativas durante o armazenamento. De acordo aos valores de BVT-N e TMA-N obtidos e aos resultados das análises microbiológicas, o músculo adutor provou ser inócua após quatro dias sob as condições de transporte e armazenagem utilizadas.

whole, fresh refrigerated, presentation for consumption on the half shell, as is the case with oysters. In practice, handlers strive to maintain the cold chain right from the moment when the specimens are harvested in their natural habitat until they are processed and/or consumed, but this is not always achieved. It is also worth to note that when delivered fresh to intermediaries or final sellers, whole organisms can fetch a price as high as 2.00 USD/scallop. It is thus necessary to look for and design alternative storage methods during transport and distribution that will assure that the quality and adequacy of the product will be maintained until offered to the final consumer. The objective of this study was to evaluate the quality and shelf life of

the adductor muscle of whole lion's paw scallops during refrigerated transport and storage, as a possible alternative for exporting the product to international markets.

### Materials and Methods

#### Experimental organisms

Adult organisms of lion's paw scallops, *N. subnodosus*, with a height of ~10cm were utilized, after being harvested from a culture system located in Bahía Tortugas, BCS, in June 2009. Before transport and storage the scallops were cleaned with an oyster machine that functions on the basis of sprayers and brushes (Ets. Bertrand, Marennes, France). The cleaning process was completed with the specimens being brushed by hand and

then cleared in 200 liter fiberglass tanks for 1h.

#### Experiment on transport and storage

The packing of the organisms prior to transport involved placing them in coolers made of expanded polystyrene with a capacity of 45 liters. The live organisms were arranged dorso-ventrally with elastic bands attached to prevent them from opening their valves and possibly drying out. A total of 120 organisms were collected and distributed into two coolers. During the packing process, the scallops were placed in a horizontal position and a bed of brown paper moistened with seawater was placed between the layers of mollusks. Once the organisms were packed, and just before seal-

ing the coolers, 5kg of frozen refrigerant gel (Dolphin Blue Crabs) at -20°C was introduced and a thermograph was placed to monitor the internal temperature of the coolers during transport, as shown in Figure 1. The refrigerant gel was separated from the upper layer of scallops by another bed of moistened brown paper. At that moment, the adductor muscles of six scallops were frozen in liquid N<sub>2</sub>, to be used to determine the initial biochemical conditions at the time of transport.

Transportation was simulated under the most realistic conditions possible. In the first phase, the coolers with the refrigerant gel and the scallops were carried in a pick-up truck at ambient temperature for 14h. Then a second stage of transportation was simulated, in which

the sealed coolers were placed in a refrigerated room at the installations of CIBNOR, La Paz, BCS. This procedure was meant to simulate the second stage of transportation, as when the organisms are transported in refrigerated trucks. The total simulated transportation time was 48h. This simulation of the stages of transportation was designed in accordance with the conditions assumed necessary to deliver this product to an international market, in this case that of the USA. In the last phase, the organisms were removed from the coolers and placed in refrigerated storage (3-6°C) in polyethylene bags to simulate the actual storage facilities utilized by the final receiver, who would then undertake to sell the product. At that moment, the adductor muscles of six organisms were dissected and frozen in liquid N<sub>2</sub> for later determination of the effects of transportation and to establish the quality and shelf life at day 0. Finally, a thermograph was placed to monitor the temperature of the refrigerated room throughout product storage (Figure 1).

#### Sampling for the study of refrigeration

The organisms transported under the simulated conditions described above remained in the refrigerated storage area for a period of four days, which is a common time to market these products. During this time, the adductor muscles of six organisms were collected, dissected, and frozen in liquid N<sub>2</sub> every 24h, and then stored at -80°C until analysis. At each point of the sampling program pH, ATP and related products, K-value, total volatile bases (TVB-N) and trimethylamine (TMA-N) were monitored. Similarly, throughout the storage period, microbiological analyses were conducted

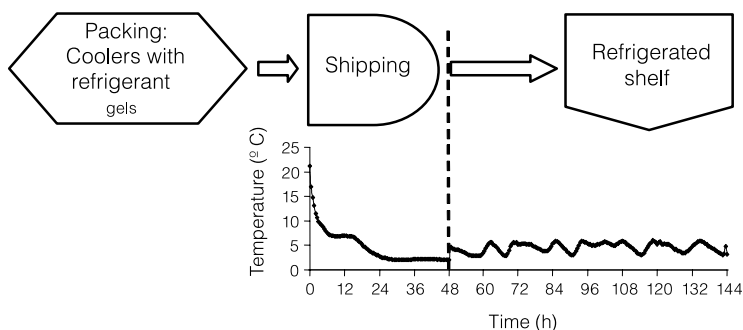


Figure 1. Preservation of organisms in refrigeration conditions, indicating temperature during the process.

in order to determine whether or not the parts were apt for human consumption. In this procedure, all of the organisms' moist tissues were extracted and homogenized together with the intervalvular liquid.

#### Proximal analysis

Quantification of moisture, proteins, lipids, carbohydrates and ash content of the adductor muscle were carried out following the methodologies described in AOAC (2005).

#### pH

The determination of pH was conducted following Woyewoda *et al.* (1986), where a homogenate of the adductor muscle was prepared with distilled water and pH was then measured using an Orion 420 potentiometer (Thermo Electron Corp.), calibrated daily using pH 4 and 7 standards as references.

#### ATP, its breakdown products, and the K-value

The extraction, identification, and quantification of the ATP (adenosine 5'triphosphate) and the products of its breakdown in the adductor muscle were conducted through high resolution liquid chromatography. The method described by Ryder (1985) was followed, where 20µl of the diluted extract were injected into a Varian Prostar 240 chromatograph (Varian Inc., Lake Forest, CA, USA) using a 4.6×150mm C18 reversed phase column (Varian)

to separate the compounds. The mobile phase used was a phosphate buffer made up of 0.04M KH<sub>2</sub>PO<sub>4</sub> and 0.06M K<sub>2</sub>HPO<sub>4</sub>. The flow rate utilized was 1ml·min<sup>-1</sup>, and detection was carried out at 254nm in a UV-Vis Varian Prostar 325 detector. The values obtained were expressed as µmol·g<sup>-1</sup> of sample. The K-value (%) was calculated according to the equation described by Saito *et al.* (1959):

$$K\text{-value (\%)} = \frac{((HxR+Hx)/(ATP+ADP+AMP+IMP+HxR+Hx)) \times 100}{}$$

where ATP: adenosine 5'triphosphate, ADP: adenosine 5'diphosphate, AMP: adenosine 5'monophosphate, IMP: inosine 5'monophosphate, HxR: inosine, and Hx: hypoxanthine.

#### TVB-N

The determination of TVB-N was conducted following the method described by Woyewoda *et al.* (1986). Here, 2g of the sample were taken and homogenized in a distilling flask with 300ml water and 2g MgO, using a T18 Basic Ultra-turrax (IKA Works, Inc, Wilmington, NC, USA). Twenty drops of vegetable oil were added as anti-foaming agent and the solution was then distilled for 25min. The distillate was collected in 15ml of 2% boric acid and then titrated with 0.05N H<sub>2</sub>SO<sub>4</sub>. A control sample was distilled under the same conditions and the values obtained were expressed as mg TVB-N/100g of sample.

#### TMA-N

The determination of TMA-N was also conducted following the method described by Woyewoda *et al.* (1986) with only slight modifications of the weights and volumes used in the extraction. The adductor muscle (3g) was homogenized in 5ml of 7.5% trichloroacetic acid in a T18 Basic Ultra-turrax (IKA Works). The homogenate was centrifuged at 3200g (4°C) in a model IEC-MULTI RF Thermo Electron refrigerated centrifuge (Thermo Fisher Scientific, Asheville, NC, USA) for 15min, and the supernatant then filtered through Whatman N° 4 paper and stored at -80°C until analysis.

For quantification, 1ml of the filtrate was taken and transferred to a screw cap test tube, diluted to 2ml with distilled water, and 0.5ml of 10% formaldehyde was added. Next, 1.5ml of 25% KOH and 5ml dry toluene were added. The tubes were shaken for 15min in a Vortex G-560 agitator (Scientific Industries Inc., Bohemia, NY, USA) and a pipette was used to remove 3.5ml of the toluene layer to be transferred to another test tube, this one with 0.3g of anhydrous Na<sub>2</sub>SO<sub>4</sub>. The mixture was shaken again until it turned transparent. Then, 2.5ml of this solution were taken and added to a test tube containing 2.5ml of a solution of 0.02% picric acid in dry toluene, which was then shaken to obtain the final solution. Measurements were conducted in a Varian UV-Visible Cary 100 Bio spectrophotometer (Varian Australia Pty Ltd.) at 410nm, using a standard curve in the range of 0.1-10mg of TMA-N. Results were expressed as mg TMA-N/100g of muscle.

#### Microbiological analyses

Microbiological analyses were carried out according to the recommended methods set out in NOM-031-SSA1-1993 (NOM, 1995) every 24h for 4 days. A total of six organisms

were collected for each 24h period. The moist tissues were extracted from the scallops and homogenized to obtain a representative sample. These procedures were conducted at the CIBNOR Microbiological Diagnostics Laboratory in La Paz, BCS, where the number of aerobic bacteria and fecal coliforms were determined in tryptone yeast extract agar media (48h at 35 ±2°C) and EC media (44 ±2°C), respectively. Also, samples were taken to be analyzed for *Vibrio cholerae* and *Salmonella* spp. (six whole organisms) for days 1 and 4 of refrigerated storage. The *V. cholerae* and *Salmonella* spp. tests were carried out in TCBS media (24h at 35-37°C) and brilliant green (24h at 35 ±0.5°C), respectively.

#### Statistical analysis

A one-way variance analysis (ANOVA) was conducted. When significant differences were found, the multiple comparison Tukey test was applied using the NCSS Ver. 2000 program (Hintze, 2001). Also, in some cases regression analyses were run. A significance level of 5% was set for all of the analyses conducted.

#### Results and Discussion

##### Proximal analysis

The percentage of moisture in the adductor muscle was 75.18 ±0.94 and was characterized by the presence of a high protein content of 17.88 ±0.46% and a low lipid content of 0.11 ±0.01%. For this reason, and following the classification reported by Domínguez and Gutiérrez (1993), the muscle was considered lean (<5% lipids). As is the case with most pectinids, *N. subnodosus* also contains considerable amounts of carbohydrates, mostly glycogen, that serve as an energy reserve in the adductor muscle; the value was 7.08 ±1.58%. Finally, the percentage of ash was

TABLE I  
VALUES OF pH, NUCLEOTIDES, NUCLEOSIDES AND BASES ( $\mu\text{mol}\cdot\text{g}^{-1}$ ), BVT-N AND TMA-N ( $\text{mg}/100$ ) OF THE LION'S PAW SCALLOP *N. subnodosus* ADDUCTOR MUSCLE TRANSPORTED AND STORED WHOLE IN REFRIGERATION CONDITIONS (3-6°C)

Parameter	Initial organisms	Pos-transport (day 0)	Storage day			
			1	2	3	4
pH	7.08 ±0.02 a	6.43 ±0.05 b	6.29 ±0.10 c	6.29 ±0.06 c	6.27 ±0.04 c	6.30 ±0.09 c
ATP	0.03 ±0.00 bd	0.02 ±0.00 cde	0.05 ±0.01 a	0.04 ±0.02 abe	0.04 ±0.01 ab	0.05 ±0.02 ab
ADP	0.33 ±0.01 a	0.25 ±0.03 b	0.26 ±0.08 b	0.23 ±0.05 b	0.21 ±0.01 b	0.18 ±0.02 b
AMP	3.95 ±0.37 a	3.61 ±0.68 a	1.44 ±0.46 b	1.08 ±0.39 b	0.99 ±0.31 b	0.80 ±0.05 b
IMP	0.05 ±0.00 b	0.08 ±0.01 a	0.12 ±0.05 a	0.15 ±0.07 a	0.13 ±0.02 a	0.14 ±0.04 a
HxR	0.10 ±0.02 e	0.76 ±0.1 d	1.43 ±0.30 c	2.00 ±0.39 ab	2.04 ±0.30 a	1.59 ±0.05 bc
Hx	0.12 ±0.03 c	0.10 ±0.09 c	0.37 ±0.07 b	0.49 ±0.07 ab	0.57 ±0.16 a	0.58 ±0.10 a
TBV-N	6.19 ±1.62 b	6.97 ±2.35 b	11.66 ±1.76 a	12.45 ±2.23 a	12.44 ±1.93 a	13.89 ±2.01 a
TMA-N	0.16 ±0.00 c	0.19 ±0.01 a	0.17 ±0.01b c	0.17 ±0.01 bc	0.17 ±0.01 b	0.17 ±0.01 b

Values are mean and standard deviation of n= 6.

Means in the same row with different superscript letters are statistically different (P<0.05).

1.40 ±0.10 and, as was the case for the values of the rest of the components, similar to that reported by Beltrán-Lugo *et al.* (2006) and Pacheco-Aguilar *et al.* (2008) for *N. subnodosus* and by Ocaño-Higuera *et al.* (2006) for the catarina scallop *Argopecten ventricosus*.

##### pH

Table I presents the data on the pH of the adductor muscle of the lion's paw scallop transported in immersion and stored whole under refrigeration. The initial value of this parameter in the organisms immediately after collection and before conditioning and storage (pH= 7.02) is similar to that reported by Kimura *et al.* (1999) for *P. yessoensis*, but higher than the value of 6.8 reported by Beltrán-Lugo *et al.* (2006), also for *N. subnodosus*. The difference between the latter figure and the initial value found in the present study may be due to the conditions employed by Beltrán-Lugo *et al.* (2006) during transport to the site where analyses were conducted, as the specimens spent 30min in a simple cooler conditioned with seawater. Under those conditions, a first stage of anaerobic glycolysis may have taken place, with the concomitant lactic acid production and pH reduction

(Durán *et al.*, 2008; Mørkøre *et al.*, 2008).

The initial neutral pH values recorded in the recently collected organisms diminished significantly (P<0.05) during the stage of simulated transport and on the first day of refrigerated storage. However, after that day and up to the end of the study, no further changes were observed. According to Riaz and Qadri (1985), an increase >0.1 units during storage indicates deterioration and loss of freshness, as occurs when alkaline-type metabolites are produced as a result of bacterial proliferation. In the present study, the pH of the adductor muscle did not increase during storage, so it can be said that in relation to this parameter the adductor muscle retained its edible quality throughout the period and under the conditions used in this study. This behavior and similar values have been reported for *N. subnodosus* and for *A. ventricosus* by Pacheco-Aguilar *et al.* (2008) and by Ocaño-Higuera *et al.* (2006), respectively.

##### ATP, Products of its Breakdown, and the K-value

Table 1 shows the values for ATP and related products obtained in the adductor muscle of the lion's paw scallop at the moment of collection, during transport in emersion,

and while stored whole in refrigeration. The first organisms obtained immediately after the cleaning process and those collected after the 48h period of simulated transport showed low levels of muscular ATP, values that may be the result of the stress generated during the conditioning and cleaning of the organisms, which fostered the breakdown of this energy metabolite. The concentration of this metabolite increased on the first day (P<0.05), but then remained with no significant variation during the rest of the storage period (P>0.05). This increase on day 1 may be due to the regeneration of ATP through the action of the adenylate kinase enzyme, or to the utilization of the phosphoarginine phosphagen in a reaction catalyzed by the arginine kinase enzyme, as has been reported for other species, such as the abalone *Haliotis discus* (Watanabe *et al.*, 1992) and the scallop *Zygochlamys patagonica* (Massa *et al.*, 2001).

The concentration of muscular ADP showed a significant variation during transport (P<0.05), but no important variations during the storage (P<0.05), whereas AMP diminished significantly (P<0.05) after day 1 of storage, but then became stable at ~1 $\mu\text{mol}\cdot\text{g}^{-1}$ , and IMP was found in concentrations  $\leq 0.15\mu\text{mol}\cdot\text{g}^{-1}$  through-

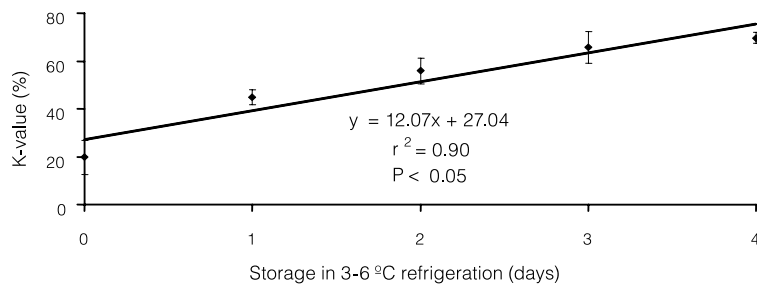


Figure 2. K-value in adductor muscle of the lion's paw scallop *N. subnodosus* stored whole in refrigeration conditions (3-6°C). Data shown are mean of n=6. Bars represent standard deviation.

out the experiment. For the latter two compounds, which are related to the sweet, pleasant flavor of seafood products, a behavior similar to that reported in *P. yessoensis* (Kawashima and Yamanaka, 1992), *Z. patagonica* (Massa *et al.*, 2001), *A. ventricosus* (Ocaño-Higuera *et al.*, 2006), and *N. subnodosus* (Pacheco-Aguilar *et al.*, 2008) was observed. On the other hand, HxR and Hx showed significant variations during the experiment, but their concentrations during the storage period were always below those reported by Ocaño-Higuera *et al.* (2006) and Pacheco-Aguilar *et al.* (2008) for *A. ventricosus* and *N. subnodosus*, respectively. This finding is relevant, since these compounds have been related to a bitter taste in seafood products. The concentrations found in this experiment could be the result of low autolytic and bacterial activity in the adductor muscle.

In the case of the K-value for the adductor muscle (Figure 2), as was to be expected in the organisms analyzed di-

rectly from the culture site, the value obtained was low,  $4.58 \pm 0.41\%$  (data not shown on the graph), although it increased significantly ( $P < 0.05$ ) during simulated transport, reaching a value of  $19.77 \pm 7.11\%$  on day 0 of the refrigerated storage experiment. This is due to the increased HxR concentration at this time.

Saito *et al.* (1959) and Sikorski *et al.* (1990) elaborated a rather general classification of the quality of seafood products with respect to their K-value. In it, a K-value  $< 20\%$  indicates a 'very fresh' product, while products with values between 20 and 50% are considered 'moderately fresh', and those  $> 70\%$  are 'not fresh'. In terms of this classification, the object of this study can be considered 'very fresh' after two days of simulated transport and, indeed, maintained that standard until the first day of refrigerated storage, when it reached  $44.87 \pm 3.24\%$ . After that, and despite the significant increase in the K-value ( $P < 0.05$ ), according to this parameter the prod-

uct remained edible until the end of the storage period. In a regression analysis, the K-value of the adductor muscle during the refrigerated storage phase showed a linear increase ( $y = 12.07x + 27.04$ ,  $r^2 = 0.90$ ,  $P < 0.05$ ). This behavior was reported previously by Ocaño-Higuera *et al.* (2006) for *A. ventricosus*.

#### TVB-N

Determining TVB-N allowed to quantify the low molecular weight volatile bases and amino compounds produced by the microbial decarboxylation of amino acids and nitrogenated compounds, which are widely used as indicators of shelf life and, more specifically, to measure stages of deterioration or loss of freshness. Table I presents the TVB-N values for the adductor muscle of lion's paw scallop *N. subnodosus* transported in immersion and stored whole in refrigerated conditions. It shows the initial low values of TVB-N ( $6.97 \pm 2.35\text{mg}/100\text{g}$ ), which are below those found in this same species by Pacheco-Aguilar *et al.* (2008) and those obtained by Ocaño-Higuera *et al.* (2006) for *A. ventricosus*. In the present study, an increase ( $P < 0.05$ ) of TVB-N was seen on day 1 of storage in refrigeration, but no additional significant change was observed ( $P < 0.05$ ). In this regard, it is important to point out that the NOM-029-SSA1-1993 norm (NOM, 1994)

establishes a maximum limit of  $30\text{mg}/100\text{g}$ . According to this parameter and our analysis, the adductor muscle was considered apt for human consumption at the end of the study in refrigeration.

#### TMA-N

One indicator that is commonly used to evaluate bacterial deterioration in seafood products is the determination of TMA-N, a compound with a characteristic ammonia odor that may reflect bacterial deterioration (Ryder *et al.*, 1984). As the results obtained from the muscle shown in Table I reveal, the initial values of TMA-N of organisms obtained directly from the lagoon increased during the transport stage, and variations were also observed during storage ( $P < 0.05$ ). However, the TMA-N concentrations found for the muscle under the conditions of transport and storage used were lower than those reported in other studies conducted with this same species and with *A. ventricosus*, even though the latter included only adductor muscles stored at a temperature of  $0^\circ\text{C}$  (Ocaño-Higuera *et al.*, 2006; Pacheco-Aguilar *et al.*, 2008). This indicator has been associated with unpleasant odors in fish and other seafood species when it exceeds certain limits. In this regard, limits of  $5\text{-}10\text{mg}/100\text{g}$  of muscle have been established (Sikorski *et al.*, 1990), which are higher than those found in the present study.

#### Microbiological Analyses

The results of the microbiological analyses carried out on lion's paw scallop during refrigerated storage, shown in Table II, reveal differences in the amount of mesophilic aerobic bacteria on different days of storage ( $P < 0.05$ ). These indexes did not reach at any moment of the experiment the maximal limit of  $5 \times 10^5$  CFU/g ( $5.69 \log_{10}$  CFU/g) allowed for human consumption according to the NOM-031-SSA1-1993

TABLE II  
MICROBIOLOGICAL ANALYSIS OF THE TOTAL WET BIOMASS  
IN THE LION'S PAW SCALLOP *N. subnodosus* TRANSPORTED AND STORED  
WHOLE IN REFRIGERATION CONDITIONS (3-6°C)

Storage time (days)	Aerobic mesophiles ( $\log_{10}$ CFU/g)*	Faecal coliforms (MPN/100 g)**	<i>Vibrio cholerae</i>	<i>Salmonella</i> spp
0	$2.42 \pm 0.08^a$	$< 30$	ND	ND
1	$3.16 \pm 0.02^b$	$< 30$	Absent	Absent
2	$4.32 \pm 0.09^d$	40	ND	ND
3	$2.24 \pm 0.09^a$	40	ND	ND
4	$3.49 \pm 0.06^c$	40	Absent	Absent

\*Values are mean and standard deviation of n=2. Means in the same column with different superscript letters are statistically different ( $P < 0.05$ ).

\*\*Statistical values calculated from Table 4 of the NMX-AA-042-1987 (NMX, 1987).

ND= Not Determined.

norm (NOM, 1994). The highest values, of 22892 CFU/g (4.32 log<sub>10</sub> CFU/g) were found on day 2 of sampling. As in the case of aerobic mesophiles, the fecal coliforms bacteria did not reach in any of the sampling days the maximum limit (230 MPN/100g), indicated by the norm. The highest values, 40 MPN/100g, were found as of day 3, but these levels were maintained until the end of the experiment. The foregoing demonstrates the innocuous nature of the product and the adequacy of the conditions employed in the handling of the organisms throughout the experiment.

Other important bacteria that must be determined and controlled according to the NOM-031-SSA1-1993 norm (NOM, 1995) are the species *V. cholerae* and *Salmonella* spp. In these two cases, a zero tolerance level is established for products of this type. The samples analyzed on day 1 and day 4 did not show the presence of these bacteria; thus, the product does not represent a risk for consumers.

## Conclusions

The biochemical, chemical, and microbiological parameters used in this study were useful in assessing the quality of the adductor muscle of lion's paw scallop *Nodipecten subnodosus* transported and stored whole in refrigeration. According to the analyses performed, the adductor muscle can be considered edible until day 4 under the conditions of transport and storage utilized in the study. These results can be achieved by maintaining the cold chain and hygienic conditions of the organisms from the moment they are harvested until their commercialization.

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