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# Changes in Quality Characteristics of Cooked and Uncooked Crab Meat (*Portunus pelagicus*) Under Ice Storage

# N. BALASARASWATHY, G. SUGUMAR<sup>\*</sup>, A. SELVAN, U. RAMESH and P.VELAYUTHAM

Department of Fish Processing Technology Fisheries College and Research Institute Tamil Nadu Veterinary and Animal Sciences University Thoothukudi - 628 008, India

# Abstract

Changes in quality characteristics of cooked and uncooked crab (*Portunus pelagicus*) meat stored directly in flake ice were determined through biochemical, microbial and organoleptic quality indices. Aerobic plate count and psychrotrophic bacterial count increased upon storage while the counts of staphylococci, total coliforms, faecal coliforms, *E. coli* and faecal streptococci remained unaltered. Human pathogenic bacteria were not detected both in uncooked and cooked crab meat throughout the storage period. Changes could be observed in the proximate composition, with increasing moisture and decreasing protein content upon ice storage. There was a decline in water extractable nitrogen,  $\infty$  - amino nitrogen and lactic acid. Glycogen level did not vary much in both cooked and uncooked crab meat upon storage while there were fluctuations in TMA and TVB contents. Organoleptic evaluation could detect changes in odour, texture and taste of crab meat with progression of storage period. Results indicated that uncooked crab could remain in acceptable quality up to 10 days while cooked crab meat could be stored up to 12 days in ice.

<sup>\*</sup> Corresponding author. Tel.: +91 461 234 0554/232 2354, Fax: +91 461 234 0574 E-mail address: sugumar\_mg@yahoo.com

# Introduction

Crab meat is an excellent source of nutrition because of its high protein component which is comparable with that of other seafoods (Srinivasagam 1979). Blue crabs are harvested from estuarine and coastal waters and are more susceptible to environmental factors and their effects on the microbiological flora of the crabs. The provisionally estimated total catch of crabs by trawlers during 2003 in India was 41,150 t of which Tamilnadu contributed the maximum (36%) to the fishery and along Thoothukudi coast, bottom - set gillnets landed 154 t of crabs during 2003 with Portunus *pelagicus* (96 t) dominating the fishery (CMFRI 2004). Crab meat is highly perishable, and unless adequate methods of preservation are adopted, the quality deteriorates rapidly. Icing is the easiest and most efficient method of preserving crabs. The high value of the product and its susceptibility to spoilage have prompted investigations into the spoilage pattern and the microbial flora responsible for spoilage (Chinnamma et al. 1970; Cockey and Chai 1991). Effect of ice and cold storage on the chemical characteristics of Egyptian crab meat (Aman et al. 1984) and changes in freshness and taste of horse-hair crab meat during ice storage (Tokunaga et al. 1983) have already been documented. This paper deals with the quality changes of cooked and uncooked crab meat under ice storage conditions.

# **Materials and Methods**

Live blue crab (*Portunus pelagicus*) of size groups in the range of 125 - 142 mm caught by bottom-set gill nets landed at the Vellapatti fishing village near Thoothukudi, Tamil Nadu were used for the study. The crabs were immediately packed in polythene bags placed in insulated boxes along with ice and transported to the laboratory. The crabs were divided into two lots, one lot was cooked in steam for 30 min and the other lot was handled without cooking. Both uncooked and cooked crabs were debacked, gutted and washed with 2 ppm chlorinated water and packed with flake ice in 1:1 ratio in thermocole (expanded polystyrene) boxes with an inner dimension of 53 x 31 x 20.5 cm and with a thickness of 2.85 cm. The crabs were buried in ice throughout the storage period with re-icing every 24 h to maintain a temperature closer to that of ice. The crabs were not individually packed so as to mimic the conditions adopted in the local crab processing industry. Samples were drawn periodically for microbi-

ological, biochemical and organoleptic assessment of quality. Microbiological analyses were performed in duplicates while biochemical and organoleptic evaluations were carried out in triplicates.

#### Microbiological evaluation

Quantitatively, the samples were analyzed for aerobic plate count (APC), psychrotrophs, staphylococci, total coliform bacteria, faecal coliform bacteria, *Escherichia coli* and faecal streptococci. Qualitative analysis was made for detection of *Salmonella, Vibrio cholerae, V. parahaemolyticus* and *Listeria monocytogenes* following standard methods (Speck 1976; FDA 1995).

The APC and psychrotrophs were enumerated by spread plating on plate count agar (Himedia, Mumbai). The inoculated plates were incubated at 5°C for 7 days for psychrotrophs while for APC, the plates were examined after 48 h of incubation at  $35^{\circ}$ C. Baird Parker agar with egg yolk emulsion and potassium tellurite was used for the staphylococci. Most probable numbers method was followed for coliform bacteria and faecal streptococci. Total coliform bacteria, faecal coliform bacteria and *E. coli* were determined using McConkey broth, Brilliant green lactose broth, EC broth and tryptone broth, while Azide dextrose broth was used for faecal streptococci.

*Vibrio cholerae* and *V. parahaemolyticus* were detected by enrichment in alkaline peptone water, selective plating on TCBS agar followed by biochemical confirmatory tests. *Salmonella* was detected by preenrichment in lactose broth, selective enrichment in tetrathionate and selenite cysteine broth followed by selective plating on bismuth sulphite agar (BSA) and xylose-lysine deoxycholate (XLD) agar. Typical colonies were subjected to biochemical tests and serological confirmation using poly "O" and poly "H" antiserum. For isolation of *Listeria*, samples were subjected to primary selective enrichment in UVM - I broth, selective enrichment in Frazer's broth, selective plating on listeria selective agar (LSA) followed by biochemical confirmatory tests.

## **Biochemical analysis**

The proximate composition of crab meat, namely moisture, protein, crude fat and ash were determined following standard methods (AOAC 1995). Glycogen (Seifer et al. 1950) and lactic acid (Jayaraman 1981) were estimated spectrophotometrically. Water extractable nitrogen was determined by Lowry's method (Lowry et al. 1951) and  $\infty$  - amino acid according to Pope and Stevens (1939). Trimethylamine (TMA) and total volatile

base (TVB - N) contents were estimated following Conway microdiffusion method (Beaty and Gibbons 1937).

#### Organoleptic evaluation

Uncooked crabs were steamed in 2% boiling brine for 10 min, the meat separated and used for organoleptic evaluation. The meat from cooked crab was used for sensory assessment after heating in steam for 2 - 3 min. Organoleptic evaluation was done by assessing various sensory characteristics such as appearance, colour, texture, odour and taste by a panel of 6 members following a 5 point scale. Scores of each attribute were pooled and the overall quality was calculated by the mean.

#### Statistical analyses

The data were subjected to statistical analysis following two-way analysis of variance (ANOVA) and correlation coefficient (Snedecor et al. 1962).

# **Results and Discussion**

#### Microbiological analysis

Quantitative microbiological changes of cooked and uncooked crab meat are shown in table 1. Aerobic plate count (APC) and psychrotrophic bacterial count of both cooked and uncooked crab meat increased with increasing time of storage. The APC increased by 1 to 2 log units in both cooked and uncooked crab meat while psychrotrophic bacteria increased from  $10^2$  to  $10^7$  cfu·g<sup>-1</sup> in 12 to 14 days of storage in ice. Psychrotrophs showed a steady increase (P < 0.01) with time of storage, while APC did not increase rapidly (P < 0.05) as the temperature of crab meat was maintained between 0.6 and 0.8°C inhibiting the mesophilic bacterial population. According to a microbiological acceptability limit of  $3.2 \times 10^6$  cfu·g<sup>-1</sup> (Villemure et al. 1986), both uncooked and cooked crab meat contained APC in levels less than the limit, although the psychrotrophic bacterial load exceeded the limit. Staphylococcal count did not increase appreciably with time of storage (P > 0.05). Fluctuations were observed in the counts of coliform bacteria and faecal streptococci. The sudden increase followed by drop in counts may be due to transient bacterial population contributed by the microbial quality of the ice used for storage, as ice was found to contain these bacteria (Table 2). Qualitative analysis of crab meat for

human pathogenic bacteria revealed absence of *Vibrio cholerae*, *V. para-haemolyticus*, *Salmonella* sp. and *Listeria* sp. both in raw and cooked crab meat throughout the study. They were also absent in the ice used for preservation. Joseph et al (1998) have already reported absence of coliform bacteria, *E. coli, Salmonella* and *Vibrio cholerae* in shrimps stored in ice. However, Rawles et al. (1995) reported that 10% of the samples of freshly cooked and picked blue crab (*Callinectes sapidus*) meat had *Listeria* sp.

storage									
Storage	G	Parameters							
days	Sample	APC	PC	SC	TC	FC	EC	FS	
0	UCC	$3.3 \times 10^3$	$<1.0 \text{ x } 10^2$	$4.0 \ge 10^2$	45.0	2.5	0.9	9.0	
	CC	$1.5 \ge 10^3$	$0.5 \ge 10^2$	$1.0 \ge 10^2$	0.9	0.0	0.0	0.3	
3	UCC	$2.5 \times 10^3$	$3.5 \times 10^2$	$2.5 \times 10^2$	4.5	2.5	0.0	15.0	
	CC	$4.0 \ge 10^3$	$8.5 \times 10^2$	$2.5 \times 10^2$	0.9	0.4	0.0	1.5	
5	UCC	$2.7 \times 10^3$	$9.0 \ge 10^2$	$1.5 \ge 10^2$	0.9	0.4	0.0	2.4	
	CC	ND	ND	ND	ND	ND	ND	ND	
7	UCC	5.9 x 10 <sup>4</sup>	$1.0 \ge 10^5$	$1.5 \ge 10^2$	0.4	0.4	0.0	5.3	
	CC	$2.3 \times 10^5$	$4.3 \times 10^5$	$2.5 \times 10^2$	0.0	0.0	0.0	1.5	
10	UCC	$1.6 \ge 10^5$	$1.5 \ge 10^6$	$2.0 \times 10^2$	0.4	0.4	0.0	4.0	
	CC	ND	ND	ND	ND	ND	ND	ND	
12	UCC	1.6 x 10 <sup>4</sup>	$1.3 \ge 10^7$	$9.0 \ge 10^2$	0.4	0.4	0.0	110.0	
	CC	$1.0 \ge 10^5$	$3.1 \ge 10^7$	$4.0 \ge 10^2$	45.0	0.7	0.3	3.0	
14	UCC	$1.0 \ge 10^4$	$7.3 \times 10^7$	$5.5 \times 10^2$	9.5	0.0	0.0	2.0	
	CC	$1.0 \times 10^5$	$4.8 \times 10^7$	$4.5 \times 10^2$	45	04	0.0	4.0	

Table 1. Quantitative microbiological changes of uncooked and cooked crab during ice storage

Legend (units): UCC: uncooked crab; CC: cooked crab; ND: not done; APC: aerobic plate count (cfu•g<sup>-1</sup>); PC: Psychrotrophs (cfu•g<sup>-1</sup>); SC: Staphylococcal count (cfu•g<sup>-1</sup>); TC: total coliforms (MPN•g<sup>-1</sup>); FC: faecal coliforms (MPN•g<sup>-1</sup>); EC: *E*.*coli* (MPN•g<sup>-1</sup>); FS: faecal streptococci (MPN•g<sup>-1</sup>)

Table 2. Bacteriological quality of flake ice

Parameter	Results
APC ( $cfu \cdot ml^{-1}$ )	$1.6 \ge 10^4$
Spore formers (cfu•ml <sup>-1</sup> )	$1.0 \ge 10^2$
Psychrophilic count (cfu•ml <sup>-1</sup> )	$1.6 \ge 10^3$
Staphylococcal count (cfu•ml <sup>-1</sup> )	Est. 4.0 x 10
Presumptive vibrio count (cfu•ml <sup>-1</sup> )	Est. 2.0 x 10
Total coliforms (MPN•100 ml <sup>-1</sup> )	95
Faecal coliforms (MPN•100 ml <sup>-1</sup> )	15
<i>E. coli</i> (MPN•100 ml <sup>-1</sup> )	7
Faecal streptococci (MPN•100 ml <sup>-1</sup> )	3
Total Clostridia (MPN•100 ml <sup>-1</sup> )	150
Salmonella (in 25 ml)	Absent
Vibrio cholerae (in 25 ml)	Absent
V. parahaemolyticus ( in 25 ml)	Absent
Listeria (in 25 ml)	Absent

# **Biochemical analyses**

The changes in proximate composition of uncooked and cooked crab meat at the beginning and termination of ice storage study are shown in table 3. Significant difference in the moisture and protein contents (P < 0.01) was observed between uncooked and cooked crab meat with lower level of moisture and higher level of protein in cooked meat than in uncooked meat. In general there was an increase in moisture content but a decrease in ash, fat and protein contents on ice storage, as already reported (Chinnamma et al. 1970; Basavakumar et al. 1998). However, there was no significant difference in the glycogen contents of uncooked crab meat samples upon storage (P > 0.05) contrary to earlier reports of decreasing glycogen content with storage time (Fatima et al. 1988). In the cooked crab meat, marked decrease was recorded as already reported. These could probably be due to individual variations in crabs.

_	Sample						
Parameters	Uncooked	l crab meat	Cooked crab meat				
-	0 day	After 10 days	0 day	After 12 days			
Moisture (%)	$79.42 \pm 0.19$	$84.77 \pm 0.7$	$76.37\pm0.25$	$82.13 \pm 0.06$			
Protein (%)	$18.01\pm0.18$	$14.13\pm0.38$	$22.55 \pm 0.15$	$16.83 \pm 0.16$			
Fat (%)	$0.61\pm0.03$	$0.28\pm0.01$	$0.51 \pm 0.03$	$0.24 \pm 0.01$			
Ash %)	$1.73\pm0.01$	$1.21 \pm 0.01$	$1.81 \pm 0.03$	$0.95 \pm 0.03$			
Glycogen (mg%)	$162.75 \pm 2.62$	$167.66 \pm 1.22$	$272.28 \pm 5.45$	$125.99 \pm 1.7$			

Table 3. Changes in proximate composition of crab meat upon ice storage

The water extractable nitrogen (WEN) content varied significantly (P < 0.01) between cooked and uncooked samples (Table 4), which might be due to denaturation and leaching of protein during cooking. There was also a significant negative correlation (P < 0.01) between storage time and WEN content of both cooked and uncooked crab meat. The WEN content decreased from 1984.52 to 1266.67 mg% in uncooked crab meat and from 854.8 to 608.2 mg% in cooked crab upon storage. Chinnamma et al. (1970) have also shown a gradual decrease of WEN with progression of storage in several crustacean and molluscan species including crabs due to continuous leaching by ice melt water.

As shown in table 4, there was a significant decrease in  $\infty$  - amino nitrogen (P < 0.05) from 160.77 to 54.67 mg% and from 113.72 to 13.2 mg% in uncooked and cooked crab meat, respectively, upon ice storage. The  $\infty$  - amino nitrogen which are formed by proteolytic activities of spoilage bacteria normally show an increasing trend upon storage, however, the decrease in its level in the present study might be due to leaching loss into ice melt water. Such declining trend in the level of  $\infty$  - amino

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Storage		Parameters							
Storage	Sample	Water extractable	α- amino nitrogen	Trimethylamine	Total volatile	Lactic acid	pН		
uays		nitrogen (mg%)	(mg%)	(mg%)	base (mg%)	(mg%)			
0	UCC	$1984.52 \pm 16.54$	$160.77 \pm 1.14$	$1.03\pm0.02$	$14.45\pm0.28$	$89.29 \pm 2.25$	$7.6\pm0.08$		
	CC	$854.80 \pm 30.61$	$113.72 \pm 0.76$	$0.34\pm0.02$	$13.59\pm0.58$	$65.00 \pm 12.48$	$7.87\pm0.10$		
3	UCC	$1971.41 \pm 14.24$	$137.37\pm0.82$	$0.97\pm0.02$	$14.58\pm0.40$	$45.66 \pm 11.87$	$7.83\pm0.07$		
	CC	$731.93 \pm 17.31$	$49.63 \pm 0.40$	UDL	$4.45 \pm 0.49$	$30.12 \pm 5.93$	$8.50\pm0.08$		
5	UCC	$1826.39 \pm 18.04$	$125.15 \pm 3.73$	$0.69\pm0.01$	$12.62 \pm 0.16$	$20.33\pm2.06$	$8.33\pm0.11$		
	CC	ND	ND	ND	ND	ND	ND		
7	UCC	$1537.35 \pm 19.98$	$67.91 \pm 1.53$	$1.02 \pm 0.03$	$8.76 \pm 0.16$	$12.90 \pm 2.28$	$8.40\pm0.09$		
	CC	$674.16 \pm 15.29$	$19.02 \pm 0.43$	$0.34\pm0.01$	$4.15 \pm 0.16$	$4.76 \pm 1.88$	$8.30\pm0.11$		
10	UCC	$1344.70 \pm 0.00$	$71.14 \pm 1.14$	$0.34\pm0.02$	$10.31 \pm 0.16$	$12.82 \pm 2.26$	$8.39\pm0.09$		
	CC	ND	ND	ND	ND	ND	ND		
12	UCC	$1310.41 \pm 22.76$	$59.41 \pm 0.64$	$0.33\pm0.01$	$17.33 \pm 1.9$	$3.02 \pm 1.28$	$8.29\pm0.07$		
	CC	$614.29 \pm 20.90$	$11.99 \pm 0.85$	$0.34\pm0.02$	$3.52 \pm 0.16$	$8.04 \pm 3.02$	$8.40\pm0.09$		
14	UCC	$1266.67 \pm 23.50$	$54.67 \pm 1.15$	$1.38\pm0.03$	$28.53 \pm 1.01$	$1.63 \pm 0.31$	$8.13\pm0.08$		
	CC	$608.2\pm10.9$	$13.2 \pm 0.91$	$0.69\pm0.01$	$4.15 \pm 0.16$	$8.4\pm2.28$	$8.3\pm0.1$		

Table 4. Biochemical changes of uncooked and cooked crab during ice storage

Legend: UCC: uncooked crab; CC: cooked crab; ND: not done

nitrogen upon ice storage of crustaceans have also been reported earlier (Chinnamma et al. 1970; Ho et al. 1986; Fatima et al. 1988; Joseph et al. 1998).

Total volatile base (TVB) and trimethylamine (TMA) are products of bacterial spoilage and their contents are often used as an index to assess the keeping quality and shelf life of seafood products (Lannelongue et al. 1982). There was no significant change in the TMA and TVB levels of both cooked and uncooked crab meat with increasing storage time in ice (P > 0.05). The TVB values of uncooked crab meat decreased from 14.45 to 8.76 mg% upon 7 days of storage and there after increased to 28.5 mg% on 14 days of storage. The limit of acceptability of seafood was reported to be 25 mg% (Lannelongue et al. 1982). The value of TVB after 12 days of storage was only 17.33 mg% well within the acceptable limit. However, the keeping quality could not be decided on these levels as there are several reports on the leaching loss leading to decreasing levels of TMA and TVB in crustaceans under ice storage (Jayaweera and Subasinghe 1988; Karthikeyan 1995).

Significant decrease (P < 0.05) of lactic acid level was observed both in uncooked and cooked crab meat with increase in storage period (Table 4). The pH values of both the samples increased slightly but not significantly (P > 0.05) with an increase in time of storage. Increase in pH level of ice stored shrimps upon storage was reported earlier (Basavakumar et al. 1988; Jayaweera and Subasinghe 1988; Chen et al. 1990).

# Organoleptic evaluation

Meat from fresh blue crabs was generally considered to have very high acceptability owing to its characteristic flavour. The organoleptic scores of uncooked and cooked crab meat on ice storage are presented in table 5. The mean score of all attributes revealing overall acceptability was 4.88 on a 5-point scale for uncooked crab at the beginning of the experiment (day 0) which reduced marginally to 4.73 on 5 days of storage in ice. Further storage resulted in loss of the characteristic odour and taste without much loss of texture properties for up to 10 days when the mean score stood at 4.05. However, on 12 days of storage, the crab meat showed distinct signs of spoilage with slight bitter to ammoniacal flavour and brown discolouration bringing down the mean score to 3.03. Using a score of 3.5 as the limit of acceptability, it was found that the uncooked crab samples remained acceptable for more than 10 days but not for 12 days of storage in ice.

Storage	Sample	Characteristics						
days		Appearance	Colour	Texture	Odour	Taste	Overall quality	
0	UCC	$5.00\pm0.00$	$5.00\pm0.00$	$4.60\pm0.47$	$4.80\pm0.24$	$5.00\pm0.00$	$4.88 \pm 0.16$	
	CC	$5.00\pm0.00$	$5.00\pm0.00$	$4.80\pm0.25$	$5.00\pm0.00$	$4.87\pm0.25$	$4.93\pm0.08$	
3	UCC	$5.00\pm0.00$	$5.00\pm0.00$	$4.17\pm0.24$	$4.67\pm0.47$	$5.00\pm0.00$	$4.77\pm0.33$	
	CC	$5.00\pm0.00$	$5.00\pm0.00$	$4.67\pm0.47$	$5.00\pm0.00$	$4.67\pm0.47$	$4.87\pm0.16$	
5	UCC	$5.00\pm0.00$	$5.00\pm0.00$	$4.00\pm0.82$	$4.67\pm0.47$	$5.00\pm0.00$	$4.73\pm0.39$	
	CC	ND	ND	ND	ND	ND	ND	
7	UCC	$4.50\pm0.50$	$5.00\pm0.00$	$3.50\pm0.50$	$4.00\pm0.00$	$4.50\pm0.50$	$4.30\pm0.51$	
	CC	$4.67\pm0.47$	$4.67\pm0.47$	$5.00\pm0.00$	$5.00\pm0.00$	$4.67\pm0.47$	$4.80\pm0.16$	
10	UCC	$4.25\pm0.25$	$4.25\pm0.25$	$4.75\pm0.25$	$3.25\pm0.75$	$3.75\pm0.25$	$4.05\pm0.51$	
	CC	ND	ND	ND	ND	ND	ND	
12	UCC	$3.75\pm0.43$	$3.75\pm0.43$	$3.00\pm0.71$	$2.38\pm0.65$	$2.25\pm0.83$	$3.03 \pm 0.64$	
	CC	$4.50\pm0.00$	$4.50\pm0.00$	$4.25\pm0.25$	$3.00\pm0.00$	$3.00\pm0.00$	$3.85 \pm 0.70$	
14	UCC	$3.30\pm0.24$	$3.00\pm0.00$	$2.80\pm0.85$	$2.20\pm0.41$	$1.70\pm0.25$	$2.60\pm0.58$	
	CC	$3.50\pm0.3$	$4.0 \pm 0.0$	$4.17\pm0.24$	$2.80\pm0.5$	$2.60\pm0.4$	$3.41 \pm 0.63$	

Table 5. Organoleptic changes of uncooked and cooked crab during ice storage

Legend: UCC: uncooked crab; CC: cooked crab; ND: not done

Description of scores:

Appearance: pieces wholesome and bright (5); not wholesome, slightly dull (4); dull (3); pieces broken (2); dry and unsightly (1) Colour: characteristic white (5); off-white to gray (4); gray to brownish (3); brownish to bluish discolouration (2); highly discoloured (1) Odour: characteristic cooked crab odour (5); loss of characteristic odour (4); odourless (3); slightly ammoniacal (2); ammoniacal, putrid (1) Taste: characteristic fresh and slightly sweetish (5); loss of characteristic taste (4); neutral or bland (3); slightly bitter (2); bitter (1) Texture: firm and elastic fibres (5); less firm (4); broken and fibrous (3); fibrous to mushy (2); pasty (1)

The cooked crab meat retained its prime quality with all original attributes intact for up to 7 days with the mean score of 4.8. After 12 days of storage loss of characteristic taste and odour were perceived however, there was not much of change in appearance, colour and texture. The score reduced to 3.85 which further dropped to 3.41 on 14 days with further loss of flavour and texture rendering the product unacceptable. On the 17th day, the product revealed strong ammoniacal odour, bitter taste and loose texture with a score of 2.75. Based on organoleptic evaluation, cooked crab meat was found acceptable for more than 12 days but not for 14 days of storage in ice. A significant negative correlation was found between storage days and over all acceptability (mean score) for uncooked (P < 0.05) and cooked crab meat (P < 0.01). Further, it was found that when the psvchrophilic bacterial count increased, the mean organoleptic score decreased significantly (P < 0.05) with an increase in time of storage for both uncooked and cooked crab. However, the shelf life of the blue crab (Callinectes sapidus) was reported to be 15.5 days at 0°C (Hong and Flick 1994), a few days higher than the finding of the present study. The exudate loss in ice melt water contributed to the loss of flavour leading to low organoleptic scores early in the storage.

# Conclusion

Upon ice storage of crab meat, the quality indices and soluble nutritive components such as  $\infty$  - amino nitrogen, TMA, TVB and lactic acid leached out in ice melt water and therefore, could not be considered reliable for assessing the shelf life of the product. Psychrotrophic bacterial load showed a steady increase of up to 7.3 x 10<sup>7</sup> and 1.0 x 10<sup>8</sup> cfu·g<sup>-1</sup> in uncooked and cooked crab meat respectively at the termination of the study. Organoleptic evaluation served as a more reliable method of assessment of keeping quality and shelf life. Based on the study, it was found that uncooked and cooked crab can be stored in ice in acceptable condition for a maximum period of 10 and 12 days respectively.

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