

Studies on the Ice Storage Characteristics of Octopus, *Octopus membranaceus* (Quoy and Gaimard)

Venkatappa and S. Dhananjaya*

Department of Fish Processing Technology,
College of Fisheries, KVAFSU, Mangalore – 575002. India

Quality changes in ice stored octopus (*Octopus membranaceus*) in different forms viz., whole octopus iced (WOI), dressed octopus iced (DOI) and dressed, packed in polythene bags & iced (DPI) were studied. During storage slight increase in moisture content was noticed in WOI and DOI samples, where as in DPI sample slight decrease was noticed. Crude protein decreased in all the samples. There was not much change in fat and ash content in any of the samples. Significant increase in TVB-N, TMA-N, PV and FFA were noticed in all the samples but the rate of change was more in DPI and DOI samples. Initial total plate count of octopus was 4.2×10^4 which increased to 9.93×10^5 , 6.50×10^5 and 8.76×10^5 cfu/g and Psychrophiles with initial count of 3.45×10^3 cfu/g increased to 3.79×10^4 , 1.90×10^4 and 2.20×10^4 in WOI, DOI and DPI samples respectively. There was decrease in nitrogenous compounds such as SSN, WSN and NPN in all the samples. It was found that the whole octopus ice stored was acceptable up to 15 days, where as DOI and DPI were acceptable up to 9 days, but DPI had little bit inferior quality than the DOI. Whole octopus iced showed significantly lower values of quality indices such as PV, FFA, TVB-N, TMA-N, TPC and psychrophilic count, indicating better quality than the DOI & DPI. Significant correlation was observed between changes in biochemical and sensory characteristics.

Key words : Octopus, Iced storage, Quality, Shelf-life

Among cephalopods, squid, cuttlefish and octopus form important marine fishery resources of India, ranking next only to finfish and shellfish (shrimps and lobsters). The estimated cephalopod landing in the year 2003-04 in India was 1,17,289 tons (CMFRI, 2003). Japan is the largest producer and consumer of cephalopods in the world; it imports about 50% of the world production. There is a potential market for octopus and has been consumed in considerable quantity.

Since Cephalopods are short lived, females reach sexual maturity shortly and produce considerable changes in muscle

proteins (Mangold, 1987). The changes in muscle protein composition may be related to changes in lipid content and moisture content. Spoilage caused by autolysis of the muscle is particularly intensive in cephalopods because of the high level of proteolytic activity in the muscle produced by their active metabolism (Stanley & Hultin, 1984), and this favours rapid microbial growth. The spoilage of cephalopods in chilled storage is governed chiefly by gram negative bacteria whereas in vacuum packed samples, aerobic organisms are inhibited and spoilage is largely dominated by gram positive bacteria (Stammen *et al.*, 1990). Application of

* Corresponding author : Ph.No.0824-2427899

high pressure is effective method of prolonging the shelf life of octopus in chilled storage, mainly due to substantial reduction of total microbial load (Hurtado *et al.*, 2002). It is important to preserve the freshness of octopus, so quick chilling or freezing of octopus and storage at low temperature will arrest the microbial growth and reduce undesirable changes in quality. Some studies pertaining to the ice-storage characteristics of squid and cuttlefish have been reported (Joseph & Perigreen, 1988; Bykovski *et al.*, 1990; Lakshmanan *et al.*, 1996; Joseph & Sherief, 2003), but data on chilled storage of octopus are very scanty. Hence, the present investigation was undertaken to study the effect of chilling by icing on the shelf-life of octopus.

Materials and methods:

Fresh samples of octopus (*Octopus membranaceus*) having 33.5 ± 4.59 cms in length and 162.8 ± 64.30 gm in weight, collected from the Mangalore landing centre were brought to the laboratory in iced condition in an insulated box. Immediately after reaching the laboratory octopus was washed thoroughly and divided into three batches. The first batch was iced in alternative layers in 1:1 ratio in an insulated box (WOI). The second batch was dressed i.e. internal organs, skin and eyes were removed and washed. The mantle and tentacles were separated and iced in alternative layers in 1:1 ratio (DOI) in an insulated box. The third batch was dressed as in the earlier case and packed in polythene bag to avoid direct contact with ice and iced (DPI) in an insulated box. Sufficient ice was covered on the top to avoid exposure to atmosphere and lid was kept closed tightly and stored at ambient conditions. During storage, ice-melt water was removed and replenished with fresh ice. The stored octopuses were sampled in triplicate once in three days for analysis.

The changes in quality were evaluated for bio-chemical parameters such as moisture, total protein and ash content (at the beginning and end of the experiment) as per AOAC (1975), total lipid was estimated according to Bligh and Dyer (1959) method. The total volatile base nitrogen (TVB-N) and tri-methyl amine nitrogen (TMA-N) were determined using the method of Beatty and Gibbons (1937). The peroxide value (PV) was estimated using the method of Jacobs (1958) and free fatty acids (FFA) using the method of Takagi *et al.* (1984). The non protein nitrogen (NPN) and water soluble nitrogen (WSN) were estimated by following the method of AOAC (1995) and salt soluble nitrogen (SSN) was estimated by following the method of Dyer *et al.* (1950).

The microbiological analyses such as total plate count (TPC) and psychrophiles were carried out as per standard methods (APHA, 1976). The sensory evaluation was carried out by trained panelists on raw and steam cooked samples on a nine point hedonic scale. Correlation coefficients (Snedecor & Cochran, 1967) between changes in biochemical and sensory characteristics during ice storage were calculated.

Results and Discussion

Fresh octopus had moisture content of 86.16%, protein content of 11.54%, lipids of 0.81% and ash content of 2.15%. Ha (1982) reported that the total lipid content of 0.5% in *Octopus vulgaris* and 0.8% in *Octopus variabilis*. During ice storage slight increase in moisture (Fig.1) was noticed in WOI and DOI samples, where as in DPI not much change was noticed. After 12 days of storage, WOI showed a higher moisture content. Similar increase in moisture content was observed by Chidanandasastri (1981) in cuttlefish stored in ice and Prafulla *et al.*, (2000) in squids stored indirectly in ice. Not

much variation in lipid content (Table 1) was observed where as a decrease was noted in protein content (Fig.2) in all the samples. This decrease can be attributed to the leaching out of water soluble proteins. Joseph *et al.* (1977) have reported a decrease in total nitrogen from 2.61% to 1.86% in squid (*Loligo* sp.) stored for 6 days

Table 1. Changes in total lipids (gm%) of octopus during ice storage.

Days Samples	0	3	6	9	12	15	18	21
	Total lipids (gm%)							
WOI	0.81	0.82	0.83	0.85	0.86	0.86	0.88	0.88
DOI	0.81	0.82	0.84	0.84	0.87	0.95	ND	ND
DPI	0.81	0.82	0.88	0.87	0.92	0.93	ND	ND

WOI = Whole octopus iced; DOI=Dressed octopus iced; DPI= Dressed and packed octopus iced

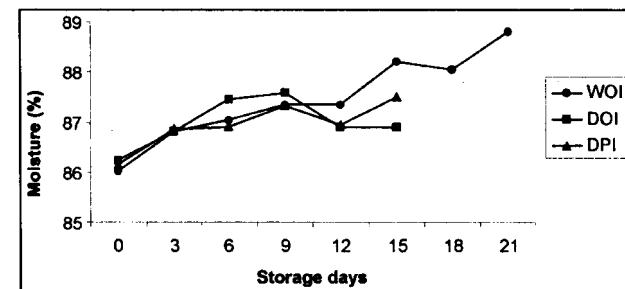


Fig.1. Changes in moisture content (%) of Octopus during ice storage.

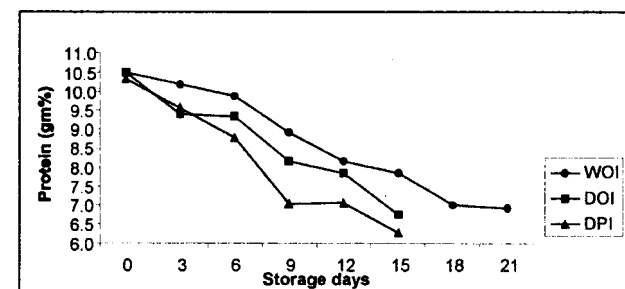


Fig.2. Changes in Protein content (%) of Octopus during ice storage.

in ice. The initial ash content of octopus was 3.56% on dry weight basis which decreased to 2.83%, 2.79% and 2.93% in WOI, DOI and DPI samples respectively at the end of storage period. Pandit & Magar (1972) reported a total ash content of 8.41 and 7.40% in case of *Sepia inermis* and *Loligo vulgaris* respectively. The differences could be mainly due to variation in species.

Changes in nitrogenous compound such as SSN, WSN and NPN of octopus during the ice storage are presented in Table-2. Salt soluble protein which occurs in high quantities in the fresh meat decreased gradually and rate of decrease was much higher in dressed samples when compared to undressed sample. A decrease in SSN of squid from 2.09gm% to 0.61 gm% of total nitrogen has been observed by Joseph *et al.* (1977) during 6days of iced storage.

Water soluble nitrogen decreased in all the samples and this could be due to leaching of water soluble proteins. According to Matsumoto (1958) 77-85% of the total protein of squid is water soluble. The ice melt water has removed most of the water soluble proteins during the iced storage. A decreasing trend of WSN of cuttlefish was also observed by Chidanandasastri (1981) from 19.04% to 10.04% of protein during ice storage period of 14 days.

Non protein nitrogen also decreased during iced storage. Joseph and Sherief (2003) have noticed similar decrease in NPN from 0.61 gm% to 0.37 gm% in cuttlefish fillets at the end of 6 days of storage in ice. Liston (1980) has suggested that some pseudomonas have very short generation times at 0 to 5 °C and are capable of utilizing NPN compounds much more rapidly as compared to other psychrotrophic bacteria.

A gradual increase in PV & FFA values

(Table-3.) were noticed in all the samples. Similar increase in PV from 7.02 millimoles of O₂/kg oil to 64.05 millimoles of O₂/kg oil was also reported by Chidanandasasthy (1981) in cuttlefish fillets stored in ice. Joseph & Sherief (2003) have noticed similar increase in PV in ice-stored cuttlefish. FFA values also showed an increased trend in all the samples during the storage period in ice. The rate of increase was more pronounced in dressed samples than the whole octopus. Joseph & Sherief (2003) have noticed similar increase in FFA of cuttlefish fillets during 6 days of iced storage.

Table 2. Changes in Salt Soluble Nitrogen, Water Soluble Nitrogen and Non Protein Nitrogen (gm%) of octopus during ice storage.

Samples	0	3	6	9	12	15	18	21
Salt Soluble Nitrogen (gm%)								
WOI	1.004	1.000	0.979	0.803	0.577	0.527	0.502	0.477
DOI	1.004	0.941	0.866	0.552	0.351	0.326	ND	ND
DPI	1.004	1.004	0.791	0.452	0.301	0.251	ND	ND
Water soluble nitrogen (gm%)								
WOI	0.412	0.392	0.261	0.251	0.241	0.221	0.211	0.201
DOI	0.412	0.321	0.231	0.221	0.221	0.161	ND	ND
DPI	0.412	0.311	0.231	0.221	0.221	0.129	ND	ND
Non protein nitrogen (gm%)								
WOI	0.306	0.261	0.241	0.241	0.223	0.181	0.161	0.120
DOI	0.306	0.221	0.221	0.201	0.181	0.090	ND	ND
DPI	0.306	0.251	0.231	0.201	0.181	0.110	ND	ND

WOI = Whole octopus iced; DOI=Dressed octopus iced; DPI= Dressed and packed octopus iced

A significant increase in TVB-N and TMA-N were observed in all the samples during ice storage (Fig.3). The rate of increase was much higher in DOI and DPI samples than WOI. Joseph & Sherief (2003) have noticed similar increase in TVB-N from 3.38mg% to 16.62 during 6 days in iced storage. Chidanandasasthy (1981) have observed an increased TVB-N content from 1.12mg% to 17.34mg% and TMA-N content from 1.83mg% to 3.05mg% in *Sepia aculeata* during 14 days of ice storage. The TMA-N content in the present experiment also

Table 3. Changes in Peroxide Value (milli moles. of O₂ / kg of fat) and Free Fatty Acids (% of Oleic acid) of octopus during ice storage.

Days	0	3	6	9	12	15	18	21
Samples	Peroxide Value							
WOI	9.03	10.13	17.88	24.62	26.67	30.87	31.13	43.43
DOI	9.03	12.11	22.86	27.90	36.12	51.27	ND	ND
DPI	9.03	11.83	32.16	28.10	36.93	53.21	ND	ND
Free Fatty Acids								
WOI	4.57	10.25	18.08	21.21	25.47	25.44	31.48	31.72
DOI	4.57	11.08	24.48	25.08	36.17	36.73	ND	ND
DPI	4.57	11.39	26.50	26.39	37.84	37.67	ND	ND

WOI = Whole octopus iced; DOI=Dressed octopus iced; DPI= Dressed and packed octopus iced

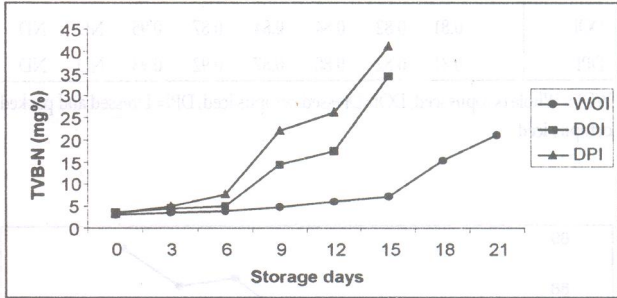


Fig. 3 Changes in Total volatile base-nitrogen (mg%) of Octopus during ice storage.

showed an increased trend in all the samples during storage (Fig.4). Woyewoda & Ke (1980) suggested 30-40mg% TVB-N and 3-10mg% TMA-N as limits of acceptability for squid. Hurtado *et al.* (2002) have observed increase in TVB-N content up to 80mg% and TMA-N content up to 25mg% in chilled stored octopus (*O. vulgaris*) by 19 days. Joseph & Sherief (2003) have noticed increase in TMA-N from 1.35mg% to 9.58mg% during 6days in iced storage. Since TMA-N production is almost entirely by bacterial action, the production of TMA-N increases with an increase in the total viable count.

In the present experiment, initial total plate count of octopus (Fig.5) was 4.2 x10⁴ which increased to 9.93x10⁵, 6.50x10⁵ and 8.76x10⁵ cfu/

g and Psychrophiles count (Fig.6) with initial count of 3.45×10^3 cfu/g increased to 3.79×10^4 , 1.90×10^4 and 2.20×10^4 in WOI, DOI and DPI respectively at the end of storage periods. Hurtado *et al.* (2002) have observed similar increase in TVC from 4 - 6 log units (log of cfu/g) in *Octopus vulgaris* during 19 days in ice storage. In the beginning of the experiment decrease in count was observed in both cases that could have been a brief latency phase,

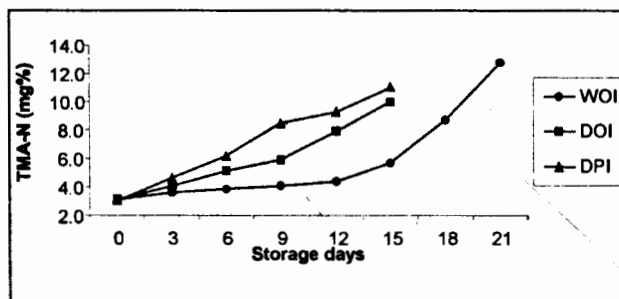


Fig. 4 Changes in Trimethylamine-nitrogen (mg%) of Octopus during ice storage.

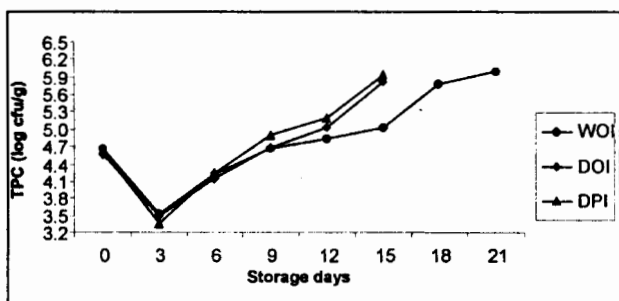


Fig. 5 Changes in Total plate count (log cfu/g) of Octopus during ice storage.

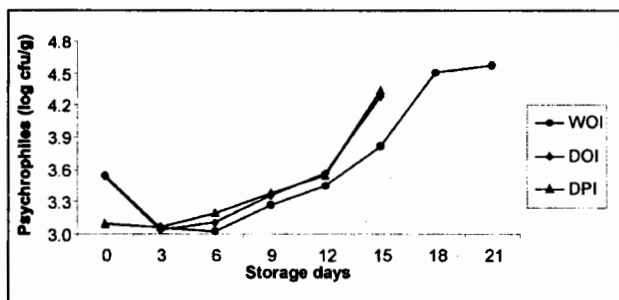


Fig. 6 Changes in Psychrophiles (log cfu/g) of Octopus during ice storage.

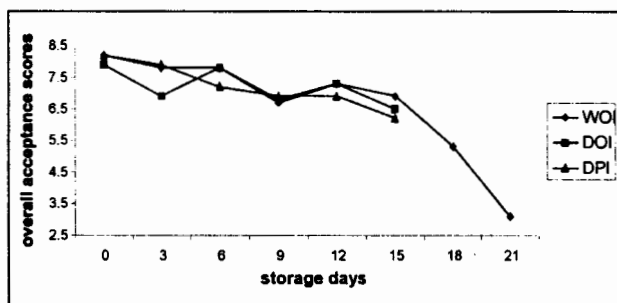


Fig. 7 Changes in Sensory scores of Octopus during ice storage.

lasting about 3-5 days. Counts were lower in whole octopus iced directly than DOI and DPI samples. The counts did not reach 10^7 cfu/g, the maximum microbiological limits set by ICMSF for foods (ICMSF, 1978) in any of the samples during the storage period. The comparatively low bacterial count may be attributed to a washing effect of ice melt water. According to Shewan (1961) it takes several days for bacteria in the viscera of ungutted fish to invade the muscle and therefore the main advantage of gutting of fish is to prevent autolytic rather than bacterial spoilage. Stansby & Lemon (1941) found that gutting of fresh mackerel could increase rather than decrease bacterial numbers. Different methods of storage affect the microbial growth. Dressed octopus lost the sensory quality much earlier than the undressed octopus. The DPI developed pink spots and lost its characteristic color on 9th day onwards, this may be due to packaging which avoided the washing effect of melted ice water. Lapa-Guimaraes *et al.*, (2002) observed more intense colour change in squids packed in polythene bags before icing and did not promote a significant reduction in microbial growth. Thus, with respect to the quality preservation of octopus under ambient conditions, non-contact ice storage method proved less advantageous when compared to contact ice storage.

Table 4. Correlation coefficient between sensory and biochemical parameters of iced octopus.

Products Parameters	WOI	DOI	DPI
PV	1.0000*	0.9352*	1.0000*
FFA	-0.6811	-0.9410*	-0.9410*
VBN	-0.9546*	-0.7134	-0.9292*
TMAN	-0.9812*	-0.6311	-0.9790*
TPC	-0.8319*	-0.8319*	-0.7683
Psychrophiles	-0.8564*	1.0000*	-0.8708*

* Significant

WOI= Whole octopus iced; DOI=Dressed octopus iced; DPI= Dressed and packed octopus iced

The mean sensory scores showed steady decrease in all the samples during the storage period in ice (Fig.7). The overall acceptance score decreased from 8.2 to 3.1 in WOI in 21 days, 8.2 to 6.5 in DOI and from 8.2 to 6.2 in DPI at the end of 15 days of storage in ice. The DOI and DPI samples retained characteristic seaweedy odour, sheen and glossy appearance, firm texture, white colour and sweet flavour up to 6 days. DPI sample had lost its flavour and characteristic colour after 9 days. Based on sensory assessment these two samples were found acceptable up to 12 days in ice, but the DPI was inferior to DOI. As the storage progressed, loss of sheen, flabby texture and discoloration with varying intensity were noticed in these samples. At the end of the storage period the colour of the meat became tinged with red. However, the WOI was found acceptable up to 15 days of storage but had fair quality at the end of 21 days of storage in ice. Good correlation coefficients (Table 4) between changes in biochemical and microbiological parameters during storage with mean sensory scores were obtained. The whole octopus lost its characteristic odor and become neutral on 15th day onwards. Studies indicated that ice storage of octopus in whole form was found better than the dressed octopus & dressed and packed condition. The reason may be that the skin of octopus might have acted as protective barrier for microbial spoilage and loss of nutritional components.

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