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Frozen Storage Characteristics of Treated and Untreated Meat from Male and Female Mussel *(Perna viridis)*

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Changes in the quality of meat obtained from male and female Indian green mussel during frozen storage are reported. The usefulness of tripolyphosphate and blanching treatment in enhancing sensory quality and in retarding progression of rancidity in frozen mussels was investigated. Changes in moisture, total protein, glycogen, trimethylamine nitrogen, total volatile base nitrogen, non-protein nitrogen, salt-soluble nitrogen, drip loss, peroxide value, total bacterial count and counts of *Escherichia coli, Salmonella* and faecal *Streptococci* in frozen meat were followed every fifteen days during storage at -18°C. The study showed that among the treated samples, male blanched mussel product could be frozen-stored in very good condition for more than 105 days and from organoleptic point of view, it had the best shelf life. However, the quality of untreated meat was acceptable for 30 days only.

Key words: Frozen storage, tripolyphosphate, blanching, quality changes, green mussel, *Perna viridis*

Indian green mussel (*Perna viridis)* is a largely unutilized source of protein. In recent years, considerable interest has emerged in developing a range of stabilized mussel products. Though mussel has immense commercial potential because of its availability from the wild as well as farmed sources, seasonal variation in quality and supply is an important drawback in its effective utilization. Ice storage characteristics of raw mussels have been reported by Chinnamma *et al.* (1970) and freezing and frozen storage characteristics have been studied by George (1974). Ablett *et al.* (1986) had undertaken a study on the influence of ascorbic acid and chelating agents on the quality of cooked mussels (*Mytilus edulis*). Jiang *et al.* (1979) worked on freezing preservation of shucked oyster (*Crassostrea gigas*) for reduction of drip loss by using pre-treatment with tripolyphosphate (1, 2 and 3%) and brine (0, 1, 2 and 3%). Boyd & Wilson (1978) have reported that meat of *M. edulis* developed strong off-flavour within two weeks of frozen storage unless adequately blanched. Seasonal changes in biochemical composition of mussels have been reported by Gabbott & Bayne (1973).

The variations that occur in the mussel during pre-spawning and post-spawning periods in male and female mussels may affect their quality during frozen storage. Information on freezing characteristics of male and female mussel meat is scanty. Moreover, there have been no reports on the comparative effect of the use of tripolyphosphate and blanching on frozen-stored mussel meat. The present investigation summarizes the results of tripolyphosphate and blanching treatment on the frozen storage quality of male and female mussel meat.

Materials and Methods

Live green mussels (*P. viridis)* were collected from Mirya creek in Ratnagiri during the second week of November, 2000. After collection, mussels were brought to the laboratory in plastic buckets with seawater. Within one hour they were cleaned with a wire brush to remove adhering epifauna and

epiflora and washed thoroughly in running seawater. They were subjected to depuration (Chellappan, 1991). The meat was removed from depurated male and female mussels and kept separately.

The shucked meat from male and female was divided into three lots each. The first lot was packed without any treatment in polyethylene bags. The second lot was packed after giving a dip treatment in 3% sodium tri-polyphosphate solution for two minutes (Heen & Karsti, 1965). The third lot was packed after blanching in a boiling solution of 2% salt for two minutes. These samples were then quick frozen at -40°C in a contact plate freezer. After freezing, the products were kept in a perforated aluminum box for cold storage at -18°C. The biochemical, microbiological and organoleptic characteristics of the meat before freezing, immediately after freezing and during storage at -18°C were determined.

Moisture, protein, fat and ash were determined by AOAC (1975) methods. Total volatile base-nitrogen (TVBN) and trimethylamine-nitrogen (TMAN) were estimated by the method of Beatty & Gibbons (1936). Glycogen content was determined by the method of De Zwaan & Zandee (1972). Non-protein nitrogen (NPN) and salt-soluble nitrogen (SSN) were estimated by the method given by Dyer *et al.* (1950). Drip loss was analyzed as per the method of Hiremath & Dhananjaya (1979). The peroxide value (PV) was determined by titrimetric method as per AOAC (1975). Total plate count of bacteria (TPC) and counts of *Escherichia coli* and faecal *Streptococci* were determined by the methods given in ISI specifications for frozen prawns (IS: 2237-1962) while *Salmonella* counts were estimated by the method of APHA (1976). Organoleptic characteristics of the products were evaluated by a trained taste panel consisting of ten members using 0-10 point hedonic scale. ANOVA (Snedecor & Cochran, 1968) was done for the overall quality characteristics with respect to frozen storage period. Least significant difference based on *t*-test was used as and when necessary.

Results and Discussion

The length and weight of whole mussels used in the present study varied from 93 mm to 104 mm and 37 g to 58 g, respectively. The average meat yield from male mussels, 29.24% and from female mussels, 38.72%. Visual inspection at the time of processing revealed that the mussels were approaching a condition of sexual maturation.

The initial moisture content in raw male and female mussel meat was estimated to be 81.7% and 82.0%, respectively. Protein, glycogen, fat and ash content in male and female raw mussel meat was found to be 58.7% and 59.0%, 18.0% and 18.3%, 10.5% and 10.8% and 5.5% and 5.6%, respectively (Table 1). These observations were in agreement with the values reported by Gopakumar (1997).

The changes in biochemical characteristics of frozen male and female mussel meat during storage at –18°C in the raw, polyphosphate treated and blanched state are presented in Table 1. Frozen storage is seen to have influenced moisture content to some extent in all the experimental samples. The extent of loss of moisture in the untreated samples was higher than in the treated ones and this was reflected in the texture characteristics as the untreated meat had lost its original elastic texture at the end of 45 days of frozen storage. These findings are in agreement with that of George (1974). The moisture content of blanched samples was significantly lower ($p<0.05$) than the polyphosphate treated samples.

The protein content of all the experimental products decreased during frozen storage period of 105 days. There were no significant differences in the level of protein in the different samples. However, a significant drop in the protein content in frozen stored raw mussels (*M. edulis*) has been reported by George (1974).

Table 1. Changes in biochemical characteristics of frozen male and female mussel meat during storage at -18°C

Characteristics	Product	Frozen storage period (days)							
		0	15	30	45	60	75	90	105
	A	80.90	79.40	78.80	77.60	–	–	–	–
	B	80.70	80.00	79.40	77.25	–	–	–	–
Moisture (%)	C	80.40	80.20	80.00	79.80	78.90	78.20	78.00	77.90
	D	80.60	80.25	79.90	79.40	79.00	78.30	78.10	77.80
	E	78.70	78.40	78.00	77.80	77.60	77.40	77.20	77.00
	F	78.60	78.50	78.10	77.75	77.50	77.35	77.10	77.05
	A	58.80	57.70	56.20	55.80	–	–	–	–
	B	58.90	57.70	56.40	56.00	–	–	–	–
Total	C	58.20	58.15	58.00	57.85	57.40	56.90	56.45	56.10
Protein (%)	D	58.30	58.10	58.05	57.75	57.50	56.80	56.40	56.00
	E	58.00	57.90	57.75	57.70	57.60	57.30	56.80	56.20
	F	58.15	58.00	57.90	57.80	57.70	57.40	56.50	55.90
	A	17.90	17.80	17.20	16.40	–	–	–	–
	B	18.10	17.90	17.10	16.50	–	–	–	–
Glycogen (%)	C	17.90	17.60	17.40	17.10	16.90	16.50	16.10	15.90
	D	17.95	17.70	17.35	17.20	17.00	16.70	16.20	16.00
	E	18.00	17.90	17.80	17.70	17.60	17.45	17.30	17.10
	F	18.20	18.00	17.90	17.80	17.70	17.55	17.50	17.00
	A	11.30	11.70	13.00	15.50	–	–	–	–
	B	11.50	11.85	13.20	15.70	–	–	–	–
TMAN (mg %)	C	11.10	11.15	11.30	11.40	11.60	12.30	12.90	13.60
	D	11.20	11.20	11.50	11.65	11.75	12.40	13.10	13.80
	E	11.00	11.10	11.20	11.30	11.50	11.80	12.20	12.50
	F	11.00	11.20	11.40	11.45	11.55	11.70	12.30	12.60
	A	8.50	10.00	12.90	15.75	–	–	–	–
	B	8.80	10.60	13.65	16.20	–	–	–	–
TVBN (mg %)	C	8.00	8.10	8.30	8.40	8.80	9.00	10.20	13.00
	D	8.10	8.20	8.40	8.45	8.70	8.90	10.50	13.50
	E	7.00	7.10	7.20	7.30	7.50	7.60	7.80	8.00
	F	7.10	7.15	7.25	7.30	7.60	7.70	7.90	8.10
	A	4.50	4.20	4.00	3.60	–	–	–	–
	B	4.55	4.00	3.80	3.55	–	–	–	–
NPN (mg %)	C	3.90	3.80	3.70	3.60	3.50	3.45	3.40	3.30
	D	4.00	3.90	3.70	3.50	3.45	3.40	3.30	3.10
	E	4.60	4.50	4.40	4.30	4.20	4.15	4.00	3.90
	F	4.70	4.60	4.40	4.30	4.20	4.15	4.05	4.00

	A	4.80	4.40	4.10	3.90	–	–	–	–
	B	4.82	4.20	4.00	3.70	–	–	–	–
SSN (mg % of TN)	C	4.10	4.00	3.90	3.80	3.75	3.60	3.50	3.30
	D	4.20	4.00	3.85	3.80	3.70	3.65	3.50	3.40
	E	5.00	5.00	4.80	4.70	4.60	4.50	4.30	4.00
	F	5.05	5.00	4.90	4.80	4.70	4.55	4.20	4.08
	A	9.20	9.70	10.00	10.40	–	–	–	–
	B	9.15	9.80	10.10	10.50	–	–	–	–
Drip loss (%)	C	7.80	8.00	8.50	9.00	9.50	9.90	10.40	10.90
	D	7.90	8.10	8,75	9.20	9.60	10.00	10.50	11.00
	E	6.85	7.00	7.50	8.00	8.40	8.80	9.20	9.80
	F	6.98	7.20	7.60	7.90	8.50	8.90	9.40	1.00
	A	3.20	4.50	5.00	5.90	–	–	–	–
PV (milli-	B	3.50	4.70	5.20	7.02	–	–	–	–
equivalents	C	3.00	3.15	3.30	3.40	4.40	5.40	6.35	7.60
$O_2.kg^{-1}$)	D	3.15	3.20	3.50	3.60	4.60	5.65	6.80	7.95
	E	2.50	2.55	2.60	2.70	2.80	2.90	3.15	3.50
	F	2.68	2.70	2.84	2.90	2.95	3.05	3.25	3.60

A - Raw male mussel meat; B - Raw female mussel meat; C - Polyphosphate treated male mussel meat; D - Polyphosphate treated female mussel meat; E Blanched male mussel meat; F = Blanched female mussel meat.

The glycogen level is higher in mussels than in other bivalve molluscs (Chinnamma *et al.*, 1970). It was higher in female meat than in male. Glycogen is the reserve material and it is stored in the connective tissue of the mantle. Immediately after spawning, the glycogen level increases gradually in all the body components (Thangavelu & Sanjeevraj, 1988). The initial level of glycogen depends on the condition of the mussel at the time of capture, i.e., the extent of struggle it undergoes before death. In all the samples, the values have shown a gradual decrease with the duration of frozen storage. A decreasing trend in glycogen content during frozen storage of edible oyster (*Crassostrea madrasensis*) meat has been reported by Balasundari *et al.* (1997).

TMAN has been found to be a useful index of spoilage and a parameter for freshness assessment of seafood. In the present study, the TMAN levels of frozen mussel meat increased during the period of frozen storage. The extent of increase was much higher in untreated samples compared to treated ones. In untreated meat, it was found to be greater than 15 mg%, the limit of acceptability, at the end of 45 days of frozen storage and developed strong off-flavour that was well correlated with the hedonic scores given by the taste panel.

TVBN content was in the range of 15-18 mg% in fresh fish, molluscs and crustaceans and it is one of the most common indices of quality used universally for deciding the state of freshness of seafood. In the present study, TVBN values were well within the limits. Similar observations are reported for cooked mussels (*M. edulis*) frozen stored at –30°C (Ablett *et al.*, 1986). The TVBN content in the blanched samples was comparatively lower than untreated and polyphosphate treated samples.

As the storage period increased there was considerable decrease in NPN values in all the samples (Table-2). This may be due to the deamination of amino acids and the

Table 2. Changes in bacterial count and overall organoleptic acceptability of frozen male and female mussel meat during storage at -18°C

Characteristics	Product Description	Frozen storage period (days)							
		0	15	30	45	60	75	90	105
TPC x 10^4 (cfu.g^{-1})	A	5.50	5.11	5.32	4.50	–	–	–	–
	B	5.10	3.80	4.22	4.80	–	–	–	–
	C	4.80	4.32	3.65	3.71	3.65	4.11	3.56	3.40
	D	4.90	4.05	3.36	4.74	4.58	4.15	3.55	3.42
	E	4.60	4.20	5.11	4.03	4.11	4.40	3.12	3.00
	F	4.40	3.34	3.80	4.38	4.32	4.20	3.02	2.90
E. coli (g^{-1})	A	175	45	52	25	–	–	–	–
	B	90	44	14	Nil	–	–	–	–
	C	115	52	57	35	18	Nil	Nil	Nil
	D	128	45	38	Nil	Nil	Nil	Nil	Nil
	E	88	32	Nil	Nil	Nil	Nil	Nil	Nil
	F	114	22	Nil	Nil	Nil	Nil	Nil	Nil
Faecal Streptococci (g^{-1})	A	135	93	29	18	–	–	–	–
	B	85	71	13	19	–	–	–	–
	C	125	113	75	32	13	Nil	Nil	Nil
	D	127	135	65	29	Nil	Nil	Nil	Nil
	E	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
	F	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Overall organoleptic acceptability (scores out of 10)	A (LSD: 0.38)	9.00	7.40	5.80	3.40	–	–	–	–
	B (LSD: 0.49)	8.90	7.40	6.10	3.60	–	–	–	–
	C (LSD: 0.32)	9.50	9.00	8.80	7.80	7.20	6.80	5.80	5.10
	D (LSD: 0.35)	9.50	9.00	8.60	8.20	7.60	6.80	6.10	5.20
	E (LSD: 0.27)	9.50	9.00	9.00	8.70	8.30	7.90	7.60	7.20
	F (LSD: 0.22)	9.50	9.00	9.00	8.65	8.30	7.80	7.35	7.15

A - Raw male mussel meat; B - Raw female mussel meat; C - Polyphosphate treated male mussel meat; D - polyphosphate treated female mussel meat; E Blanched male mussel meat; F = Blanched female mussel meat.

possible losses through thaw drip. All the experimental products had an initial NPN content of around 4 mg%, which might have originally formed in the raw material due to autolytic action. The SSN content in frozen mussel meat decreased during storage. The extent of decrease was greater in untreated samples compared to treated samples. The SSN content in untreated and polyphosphate treated samples decreased sharply compared to blanched samples at the end of frozen storage. This was reflected in the flavour scores and in the organoleptic rating. Similar decreasing trends have been noted in raw

mussels (*M. edulis*) and clams (*Villorita* spp.) by George (1974) and in edible oyster (*Crassostrea* spp.) by Sawant (1997) during frozen storage.

The extent of drip loss was observed to be significantly higher ($p<0.05$) in untreated samples, and among the treated samples, it was much less in blanched samples than the polyphosphate treated ones. Duration and temperature of storage were the important factors influencing the drip loss. George (1974) reported that in the case of raw mussels and clams, the amount of free drip increased with increase in storage period and that the drip characteristics are directly related to the quality of product. Balasundari (1997) observed similar trend in drip loss as was found in the present study. In order to reduce drip losses, Jiang *et al.* (1979) had tried some special immersion pre-treatments for oysters (*C. gigas*). These included brining, treatment with sugar and immersion in sodium polyphosphate solution. Jiang *et al.* (1985) concluded that the combined use of immersion and ice glaze treatments was effective for preventing discolouration and reducing drip loss in frozen storage of *C. gigas*.

The changes in PV (Table 2) indicated that lipid oxidation occurred in all the samples. After 30 days, PV for untreated and polyphosphate treated samples had increased sharply compared to initial levels ($p<0.05$). A definite effect of blanching could be observed as the PV was comparatively low in the blanched samples and it was reflected in all the organoleptic attributes, especially the flavour. It was also observed that meat from female had significantly higher ($p<0.05$) PV compared to meat from male animals. Jeong *et al.*, (1991) had observed that in oysters, PV increased gradually with the duration of storage. Similar observations have been reported by Sawant (1997). Ablett *et al.* (1986) reported that lipid oxidation occurred in frozen cooked mussel, particularly under marginal frozen storage temperatures.

The total plate count showed considerable variation during cold storage of all the products (Table 2). Among the products examined, the TPC was in the order of raw meat > polyphosphate treated meat > blanched meat. Reduction in counts of organisms like *E. coli* and faecal *Streptococci* was observed during frozen storage and were completely destroyed at the end of 75 days. This was in accordance with the findings of George (1974). However, the counts of these organisms were observed to be nil in blanched products. *Salmonella* was not detected in any of the samples. The bacteriological quality of treated products was found to conform to the specifications laid down by Export Inspection Agency (EIA, 1995).

The shelf life with respect to organoleptic quality of experimental samples indicated that all the characteristics decreased gradually during frozen storage (Table 2). However, in untreated mussel meat samples, all the organoleptic characteristics, especially the texture and odour, decreased sharply. The overall scores were beyond the acceptable limits (5.0) at the end of 45 days of frozen storage. The organoleptic changes observed in the untreated mussel meats were mainly change of colour to dull white and finally to brown in spoiled samples, loss of firmness and elasticity of the meat, development of sponginess, breaking up of the flesh and resultant oozing out of drip and loss of characteristic flavour.

The quality of male and female untreated mussel meat was acceptable up to 30 days only, while overall acceptability of polyphosphate treated and blanched samples indicated that the quality of the product was acceptable for up to 105 days and more than 105 days, respectively. From the organoleptic point of view, male blanched mussel product had the best shelf life.

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