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Effect of Pre-chilling on the Shelf-life and Quality of Silver Pomfret (*Pampus argenteus*) Stored in Dry Ice and Wet Ice

G. Jeyasekaran, P. Ganesan, R. Anandaraj, R. Jeya Shakila and D. Sukumar
Department of Fish Processing Technology, Fisheries College and Research Institute, Tamilnadu Veterinary and Animal Sciences University, Tuticorin 628 008, India

Abstract: Effect of pre-chilling on the sensory, microbiological and biochemical quality of silver pomfret (*Pampus argenteus*) stored in a standardized combination of dry ice and wet ice at the ratio of 1:0.2:0.5 (wt/wt/wt) was studied. Silver pomfret were chilled in ice till their core temperature reached to 0°C before their storage in the combination of dry ice and wet ice. The pre-chilled fish stored in the combination were sensorially acceptable up to 27 h without reicing, whereas the fish was acceptable only up to 18 h when they were stored without pre-chilling. Total bacterial count, psychrophilic count, H_2S producers and lactics count was 10^7, 10^6, 10^5 and 10^3 cfu g^{-1}, respectively at the end of storage period in the pre-chilled fish. *Moraxella* constituted 50% of the total bacterial flora in raw pomfret and became 2%, when they were pre-chilled with ice. *Moraxella* was also found to be the dominant (29%) flora in pre-chilled stored pomfret, whereas *Flavobacterium* was dominant in control pack. TMA-N and TVB-N did not exceed the limit of acceptability. Hypoxanthine (Hx) and Free Fatty Acids (FFA) content were 7.94 mg/100 g and 19.84% as oleic acid, respectively at the end of the storage period.

Key words: Pre-chilling, dry ice, wet ice, bacteriology, biochemical, sensory

Introduction

The rate of deterioration in fish is highly temperature dependent and can be inhibited by the use of low storage temperature (Sivertsvik *et al*., 2002). Chilling generally slows down the deterioration of seafood. The prevalent method of retarding spoilage of seafood in India, as well as in other tropical countries, is storage in ice (Surendran *et al*., 1989). The most common chilling medium for preserving fresh fish is ice. However, the quantity of crushed ice required for chilling fresh fish is quite substantial which is at least 1:1 ratio (wt/wt) and sometimes is even higher with tropical conditions (Lima dos Santos *et al*., 1981). When the atmospheric surrounding of the product is modified to reduce oxygen concentration, the shelf life is increased considerably due to the reduction in the rate of chemical oxidation by oxygen as well as in the growth of aerobic microorganisms (Stiles, 1991).

Carbon dioxide is the most important gas used in modified atmospheric packaging of fish. Because of its bacteriostatic and fungistatic effect, it inhibits the growth of many spoilage bacteria (Sivertsvik *et al*., 2002). Carbon dioxide enriched atmospheres have been increasingly used in the seafood trade during the last few years (De la Hoz *et al*., 2000). Dry ice has gained popularity in India for the rapid transportation of fresh fish by air by reducing the temperature rapidly as well as modifying the packing environment with CO_2. It acts as coolant in the present trends of shipping of fresh seafood (Schoemaker, 1990). Recently, dry ice is often mixed with wet ice to save shipping weight, cost and extend the cooling energy of wet ice. Several exporters use dry ice blindly in combination with wet ice without any scientific basis for transportation of fresh fish in chilled

Corresponding Author: G. Jeyasekaran, Department of Fish Processing Technology, Fisheries College and Research Institute, Tamilnadu Veterinary and Animal Sciences University, Tuticorin 628 008, India, Tel: +91 461 2322354 2340554 Fax: +91 461 2340574 2340401

condition. It has been reported by us that the fish, Emperor breams (*Lethrinus miniatus*) stored in a combination of dry ice and ice at the ratio of 1:0.2:0.5 (wt/wt/wt) were superior when compared to their storage in wet ice only (Jeyasekaran *et al*., 2004a). However, there is no clear report on the effect of pre-chilling on the quality of fish stored in dry ice and wet ice. Since silver pomret (*Pampus argenteus*) is one of the commercially important fish varieties that is chilled and exported from India and the knowledge of specific spoilage organisms of different fish from various aquatic environments of tropical region under different packaging condition is still limited (Sivertsvik *et al*., 2002), the present study was undertaken to investigate the effect of pre-chilling on the quality of silver pomfret (*P. argenteus*) stored in the standardized combination of dry ice and wet ice in relation to their shelf life.

Materials and Methods

Raw Material

Silver pomfret (*Pampus argenteus*) were procured from fish landing center of Tuticorin, India, which is situated in the same campus where our laboratory is located. Time interval between harvesting and the arrival of fish at the landing center was 6 to 7 h and during this period they were iced. They were immediately brought to the laboratory in insulated containers. They had an average length of 24 cm and weight of 195 g. Whole fish were beheaded, gutted and washed in tap water. They were divided into 2 lots of 15 kg each. First lot was chilled with wet ice till the core temperature of fish reached to 0°C, which took 40 min. Pre-chilled fish were packed with a standardized combination of dry ice (Thermosafe Dry ice Machine, USA) and wet ice (Ziegra Flake ice Maker, Germany) at the ratio 1: 0.2: 0.5 (wt/wt/wt) of fish to dry ice to wet ice (Jeyasekaran *et al*., 2006). Second lot was packed with the same combination of dry ice and wet ice without pre-chilling, which served as control. Gloves were worn during handling of ice and fish. Care was taken to avoid direct contact of fish with wet ice and dry ice. In order to avoid direct contact between the ice and fish, dry ice was wrapped in kraft paper pouches and wet ice in polythene bags. Fish was not re-iced during the entire process of study. Packages were wrapped in polythene bags (Polypropylene, 200 gauge), placed in conical shaped styrofoam boxes and sealed airtight with cellophane tape. The boxes were stored at room temperature (33±1°C). One pack from each lot was periodically opened and analyzed in triplicate for sensory, bacteriological, biochemical and physical quality until sensory rejection. This study was conducted during the months of August - October 2005 in the Fish Quality Control Laboratory of Department of Fish Processing Technology, Fisheries College and Research Institute, Tuticorin, India.

Sensory Evaluation

Sensory characteristics and overall acceptability of silver pomfret (*P. argenteus*) were assessed by a panel of six experienced Faculty of Fisheries College and Research Institute on the basis of ten point scale on each sampling. Sensory characteristics studied included general appearance (inclusive of color), odor and texture of fish. Scale employed for evaluating sensory quality of chilled silver pomfret was developed based on the guidelines given by Lima dos Santos *et al*. (1981) and is given in Table 1. The scores were given in the decreasing order scale with 10-9 for excellent, 8-7 for good, 6-5 for fair and acceptable, 4-3 for poor and 2-1 for very poor. The mean of the scores given by the panel represented the overall sensory quality (Huss, 1988). A score of 3 to 4 constituted unacceptable and shelf life failure.

Bacteriological Quality Analysis

Bacteriological analysis carried out in this study included total bacterial count, total psychrophiles, total lactics, total H_2S producers, total coliforms and total anaerobic sulphite reducers. Fish muscle (25 g) was homogenized using 225 mL physiological saline (0.85% NaCl) and serial

Table 1: Scoring system for evaluating sensory quality of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

General appearance	Texture	Odor	Score
Bright opalescent sheen, cornea transparent, eyes perfectly fresh, convex black pupil, bright red gills, slime transparent, no bleaching	Very firm, elastic to finger touch, scales very firmly attached to skin	Fresh seaweedy odor	10
Eye fresh, slightly convex, black pupil, red gills, slime translucent, cornea translucent	Moderately firm, elastic	Loss of fresh seaweedyness, shellfish odor	9
Eyes flat, very slight gray pupil, dull red gills, slightly translucent cornea	Firm, moderately elastic, scales firmly attached to skin	No odors, neutral odor	8
Slime translucent, eyes flat, slight gray pupil, loss in red color of gills	Slightly firm, slightly elastic	Slightly musty, mousy, milky, garlic or peppery odor	7
Eyes slightly sunken, gray pupil, slight opalescent cornea, discoloration of gills, some mucus, outer slime slightly opaque	Slight soft, some grittiness near tail	Bready, malty, beery, yeasty odor	6
Eyes sunken, pale pupil, opaque cornea, slime opaque, some mucus, light brown gills	Moderately soft, moderate grittiness, slightly loose scales	Lactic acid, sour milk or oily odor	5
Eyes completely sunken, milky white pupil, opaque cornea, brown gills	Soft, definite grittiness, slightly loose flesh, scales easily removable	Acetic or butyric acid, grassy, slightly sweet, sweaty or chloroform odor	4
Eyes completely concave, head shrunken with thick slime, gills exhibit bleaching and dark brown discoloration	Very soft, marked grittiness, loosened flesh, scales easily rubbed off the skin	Stale cabbage water, wet matches, phosphine like odor	3
Eyes completely concave, shrunken head and body, cornea and pupil milky white, body covered with yellowish mucus or slime	Very soft and flabby, slight retaining of finger indentation, flesh easily torn	Ammonical with strong odors	2
Eyes loose and completely concave, body and head shrunken and discolored, bloom completely gone, thick yellowish slime or mucus	Extremely soft and flabby, strong retention of marks, flesh very easily torn	Indole, fecal, H_2S, strong ammonical and putrid odors	1

decimal dilutions of each homogenate were carried out with the same diluent for the respective bacteriological analysis (APHA, 2001). Appropriate dilutions were spread plated onto Trypticase Soya Agar (TSA) for the enumeration of total bacterial count and total psychrophilic count. The plates were incubated at 37°C for 24 h for the enumeration of total bacterial count, whereas they were incubated under refrigerated condition (5°C) for 7 days for the enumeration of psychrophiles. The colonies from mesophilic plates (having 30 to 300 colonies) at each sampling were isolated and identified by various biochemical tests (LeChevallier *et al*., 1980; Balows *et al*., 1992). Double-layer pour plate technique was followed for the enumeration of total lactics using deMan Rogosa Sharpe (MRS) lactobacillus agar (Surendran *et al*., 2003). Pour plate technique was followed for the enumeration of H_2S producers using peptone iron agar. Inoculated plates were incubated at the room temperature for 48 h. After appropriate incubation, the number of colonies developed on the plates were counted and expressed as colony forming units per gram (cfu g^{-1}). Most Probable Number (MPN) technique was followed for the enumeration of total coliforms and total anaerobic sulphite reducers using lauryl sulphate tryptose broth and Differential Reinforced Clostridial Medium (DRCM), respectively. The tubes were incubated at 37°C for 24 h for the enumeration of total coliforms. The tubes showing acid and gas production were counted as positive and expressed as MPN count g^{-1}. The inoculated tubes were incubated in a water bath at 37°C for 4 days for anaerobic sulphite reducers. The tubes exhibiting black precipitate were counted as positive and expressed as MPN count g^{-1}.

Biochemical Quality Analysis

Biochemical quality indices studied included Total Volatile Base Nitrogen (TVB-N), trimethylamine nitrogen (TMA-N), hypoxanthine (Hx) and Free Fatty Acid (FFA). TVB-N and

TMA-N contents were determined by the Conway micro-diffusion method (Cobb *et al*., 1973). Hx content was estimated using the method of Burt (1976). Free fatty acid content was determined by the method described by Pearson (1968).

Physical Quality Analysis

Physical parameters studied were pH, temperature and gas composition. pH was determined by using pH meter (Digisun Electronics Digital pH Meter 707, India) by taking 10 g of homogenized sample in 100 mL distilled water. Changes in temperature of packages and storage room were recorded by using Ultrafreezer temperature probe (Consort Model T 852, Belgium). Gas composition of packages and storage environment was measured by gas analyzer (PBI Dansensor CheckMate 9900, Denmark).

Statistical Analysis

Analysis of variance (ANOVA) was performed by using statistical package (SPSS 10.0) to examine whether any significant difference exists between different treatments, with respect to the different fish quality characteristics at 5% level.

Results and Discussion

Sensory Quality

Changes in the sensory scores of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 1. Raw fish exhibited fresh seaweedy odor, bluish-white translucent appearance and very firm texture with an overall sensory score of 9.8. Initial characteristic features were maintained until 12 h of storage in pre-chilled package, whereas fish in control pack retained the initial characters only up to 1 h of storage. On 15 h, pre-chilled fish lost its fresh seaweedy odor, but appeared as shiny white with firm texture, whereas fish in control pack exhibited slight ammoniacal odor. Even on 21 h, fish in pre-chilled package exhibited shiny appearance, but the skin became very slight yellowish in color, whereas in control pack, the fish were sensorially unacceptable with strong fecal odor and complete loss of texture with a score of 3.9. On 27 h, fish slightly lost its texture, meat color became slight red and exhibited ammoniacal odor. Huss *et al*. (1997) reported that the release of off-odors might be due to chemical changes in fish muscle. The fish became sensorially unacceptable on 30 h with strong ammoniacal odor and complete loss of texture. The changes in the sensory score between the treatments were found to be significant ($p<0.05$). It has been earlier reported that

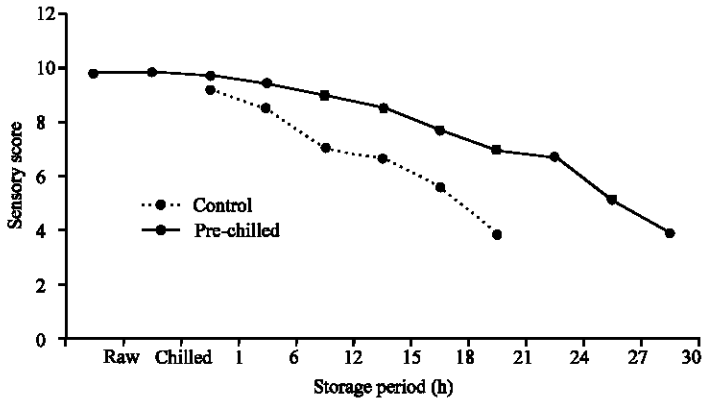

Fig. 1: Changes in the sensory score of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

combination of dry ice and wet ice at this ratio (1:0.2:0.5) extended the storage life of seerfish (*Scomberomorus commersonii*) by about 60% when compared to ice alone (Sasi *et al*., 2003).

Bacteriological Quality

Changes in total bacterial count of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 2. The initial total bacterial count was found to be 10^5 cfu g^{-1}, which was maintained when the fish were chilled with ice before packing. On 1 h of storage, total bacterial count reduced by a log, which then increased only after 18 h in pre-chilled pack. But, in control pack, the initial count reduced by a log only on 6 h and the same was maintained up to 12 h of storage. The changes in total bacterial counts between the two treatments were statistically significant ($p<0.05$). Earlier workers also reported that the application of CO_2 gaseous environment and exposure to low temperature inhibited the bacterial growth (Clark and Lentz, 1969; Jay, 1987). On further storage, total bacterial count increased continuously and reached a count of 10^7 cfu g^{-1} at the end of storage period. Papadopoulos *et al*. (2003) also observed that a mesophilic count of 10^7 cfu g^{-1} was the maximum level for the acceptability of marine fish.

Bacterial flora associated with raw fish, chilled fish, pre-chilled and dry ice cum wet ice stored fish and control pack fish stored in a combination of dry ice and wet ice are shown in Fig. 3. Fresh fish carried a bacterial flora comprising of *Moraxella, Micrococcus, Alcaligenes, Pseudomonas* and *Corynebacterium*. *Moraxella* constituted 50% of the total bacterial flora. Shift in bacterial flora was observed when the fish were chilled with ice before packing. *Corynebacterium* constituted about 31% of the flora and *Aeromonas* and *Escherichia coli* newly appeared in chilled fish in addition to the flora already observed in the raw fish except *Pseudomonas*. *Moraxella* (29%) again dominated when the pre-chilled fish were stored in a combination of dry ice and wet ice. It was followed by *Pseudomonas, Aeromonas*, *Alcaligenes*, *Micrococcus*, *Escherichia coli*, *Flavobacterium*, *Corynebacterium* and *Plesiomonas*. *Pseudomonas* and H_2S producing bacterial population were found to be specific spoilage bacteria in fish from tropical waters (Gram and Huss, 1996). However, Lindsay *et al*. (1986) reported that dominant spoilage flora such as *Pseudomonas* and *Shewanella* in fish stored under aerobic refrigerated conditions were effectively inhibited by atmosphere enriched with 20% CO_2 or more and this might be the reason, *Pseudomonas* constituted only 17% of the flora in the present study. *Flavobacterium* (36%) was dominated in control pack fish followed by *Acinetobacter*, *Pseudomonas*, *Alcaligenes*, *Bacillus*, *Staphylococcus*, *Moraxella*, *Micrococcus* and *Vibrio*.

Changes in total psychrophiles count of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 4. Fresh fish exhibited a psychrophilic population of 10^4 cfu g^{-1}, which

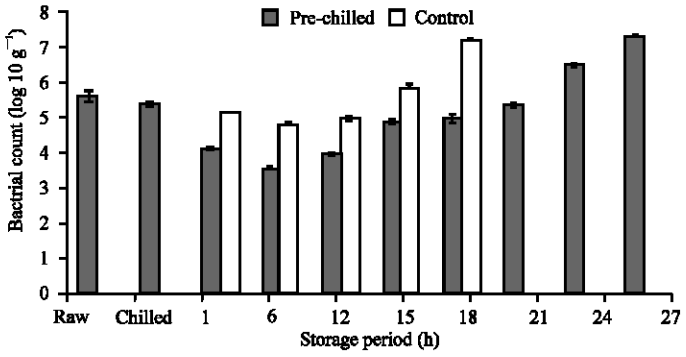

Fig. 2: Changes in total bacterial count of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

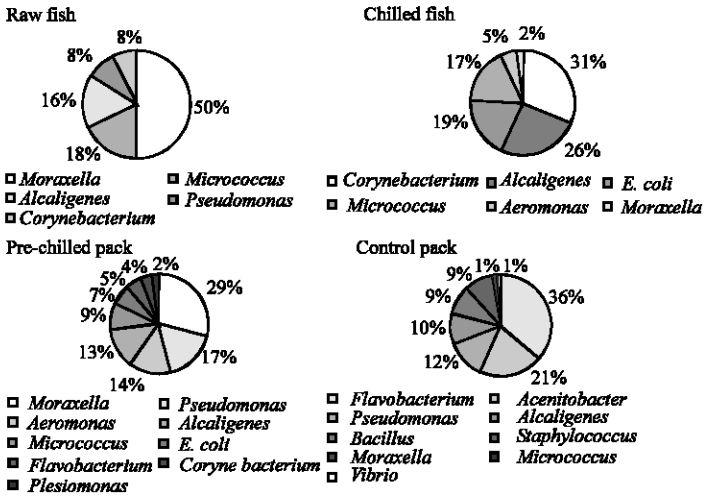

Fig. 3: Bacterial flora associated with raw, chilled, fish in pre-chilled and control pack stored in a combination of dry ice and wet ice

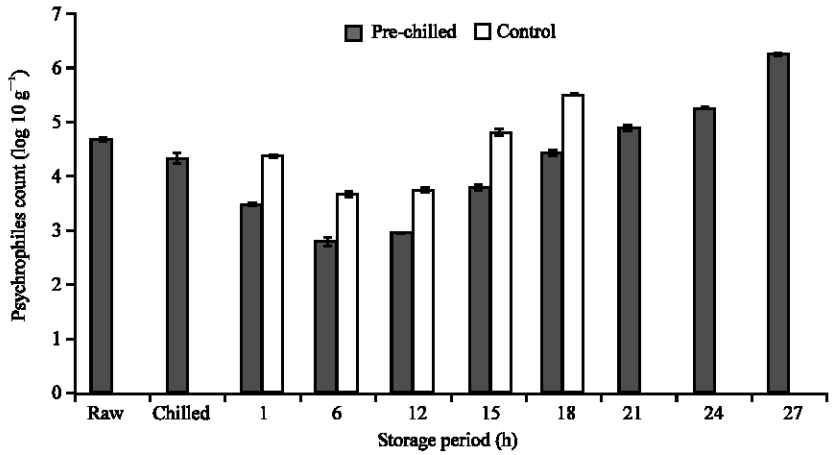

Fig. 4: Changes in total psychrophilic count of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

maintained when the fish were chilled with ice before packing and also on the 1 h in control pack. In pre-chilled pack, psychrophilic population reduced by a log on the 1 h and further reduced by another log on the 6 h and the same was maintained until the 12 h of storage. The population continuously increased from 15 h onwards and reached to 10^6 cfu g^{-1} at the end of storage period. Similar psychrophilic count of 10^6 cfu g^{-1} was also observed by Jeyasekaran *et al*. (2006) when tuna (*Euthynnus affinis*) chunks packed in a combination of dry ice and wet ice. However, in the control pack, the initial population reduced by a log on the 6 h, which maintained up to 12 h. On further storage, the population increased and reached to 10^5 cfu g^{-1}. Changes in psychrophilic count showed significant difference ($p<0.05$) between the treatments.

Changes in total lactics count of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 5. Fresh fish had a total lactics count of 10^2 cfu g^{-1}, which maintained until the 21 h of storage. Similar initial lactics count was also found in our earlier studies in tuna

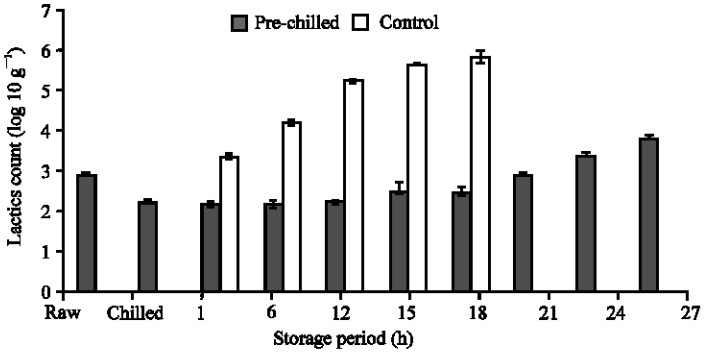

Fig. 5: Changes in total lactics count of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

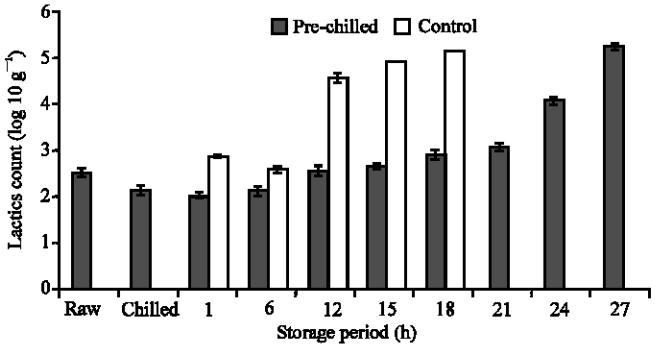

Fig. 6: Changes in total H_2S producers count of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

(*Euthynnus affinis*) chunks stored in the combined package of ice (Jeyasekaran *et al*., 2006). On the 24 h, lactics count increased by a log and the same was maintained until the end of storage period. Jay (1987) reported that most of the lactic acid bacteria could grow in the pH range of 4.0 to 4.5. Since the pH of silver pomfret varied from 6.23 to 6.95 in the present study, the growth of lactics was not high. In the case of control pack, initial count increased by a log on the 1 h and further to 10^5 cfu g^{-1} on the 12 h, which maintained till the end of storage period. The changes in total lactics count between the treatments were statistically significant ($p<0.05$).

Changes in total H_2S producers count of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 6. Initial H_2S producers count was 10^2 cfu g^{-1}, which maintained till the 18 and 6 h in pre-chilled and control pack, respectively. Huss (1988) suggested that CO_2 has an inhibitory effect on H_2S producing bacteria and their count was comparatively lower than the total bacterial load, which supported the findings of the present investigation. On further storage, H_2S producers increased by a log at each sampling period from 21 h onwards to reach 10^5 cfu g^{-1} at the end of storage period in pre-chilled fish, whereas in control pack the initial count increased to 10^4 cfu g^{-1} on the 12 h, which maintained up to 15 h and reached 10^5 cfu g^{-1} at the end of storage period. Significant difference ($p<0.05$) in H_2S producers count was observed between the treatments.

Changes in total coliforms and anaerobic sulphite reducers count of pre-chilled pomfret stored in a combination of dry ice and wet ice are presented in Table 2. Initial coliform count was found to be 13 MPN g^{-1}, which reduced to 8.0 MPN g^{-1} when the fish chilled before packing. In pre-chilled pack, the coliforms population became zero on the 1 h, which maintained up to 6 h, whereas in control pack, the count became zero only on the 6 h of storage. Ramachandran *et al*. (1990) found the

Table 2: Changes in total coliforms and total anaerobic sulphite reducers of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

Storage period (h)	Total coliforms (MPN g^{-1})		Total anaerobic sulphite reducers (MPN g^{-1})	
	Pre-chilled	Control	Pre-chilled	Control
Raw	13	13	0.9	0.9
Chilled	8.0	-	0.9	-
1	0	5.0	4.5	4.5
6	0	0	9.5	9.5
12	7.0	345	7.5	110
15	8.0	920	0	140
18	14	1600	0	140
21	175	DC	0	DC
24	212		0	
27	345		0.4	

DC-Discontinued

presence of coliforms particularly *E. coli* in immediately iced Chinese herring, *Hilsa toli* at a level of 260 MPN g^{-1}; however, it did not survive after 2 days of storage. Coliforms reappeared on the 12 h in both the packs and on further storage, the count increased gradually to 345 MPN g^{-1} in pre-chilled pack and 1600 MPN g^{-1} in control pack at the end of storage period. The initial total anaerobic sulphite reducers were 0.9 MPN g^{-1}. The same level of anaerobic sulphite reducers count was also initially observed by Jeyasekaran *et al*. (2004b) in tuna (*Euthynnus affinis*). On the 1 h of storage, the count increased to 4.5 MPN g^{-1} and then to 9.5 MPN g^{-1} on the 6 h of storage in both the packages. This might be due to the displacement of oxygen by carbon dioxide inside the package. On further storage, fish in control pack exhibited higher count of 140 MPN g^{-1}. But, count in pre-chilled pack became zero on the 15 h onwards to 24 h of storage.

Biochemical Quality

Changes in TVB-N content of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 7. Fresh fish had a TVB-N content of 5.19 mg%, which decreased after chilling and further to 3.79 mg% during storage on 1 h, whereas in control pack, TVB-N content increased to 12.45 mg% on the 1 h. On further storage, TVB-N content was found to be in an increasing trend in both the packs and exceeded the acceptable limit in control pack at the final storage period. Significant difference ($p<0.05$) was observed in TVB-N contents between the treatments. Similar increasing trend was also reported in our earlier studies in emperor breams (Jeyasekaran *et al*., 2004a). Leblanc and Leblanc (1992) also found that haddock (*Melanogrammus aeglefinus*) fillets super-chilled with CO_2 snow exhibited lower TVB-N content than iced fillets.

Changes in TMA-N content of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 8. Initial TMA-N content of fresh fish was 2.72 mg%, which decreased after chilling with wet ice and further reduced to 1.32 and 2.50 mg% on the 1 h of storage in pre-chilled and control pack, respectively. On further storage, TMA-N contents were found to increase gradually, but did not exceed the acceptable limit in pre-chilled pack at the final storage period, which might be due to the effect of CO_2 on TMO reducing bacteria (Sasi *et al*., 2003). Changes in the TMA-N contents between the treatments are statistically significant ($p<0.05$). It has also been reported that the rejection limit for TVB-N and TMA-N content in fish varied with species and processing condition (Huss, 1988; Dalgaard, 2000).

Changes in hypoxanthine (Hx) content of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 9. Fresh fish had an initial hypoxanthine content of 3.26 mg/100 g, which decreased to 2.90 mg/100 g after chilling and further reduced to 2.48 mg/100 g on the 1 h in pre-chilled pack, whereas the content was 3.40 mg/100 g in control pack. Hypoxanthine content was in increasing trend during further storage and reached to 7.94 and 8.88 mg/100 g in pre-chilled and control pack, respectively at the end of storage period. The changes in hypoxanthine content between the treatments were found to be significant ($p<0.05$). Our earlier study on lethrinus (*Lethrinus miniatus*) stored in

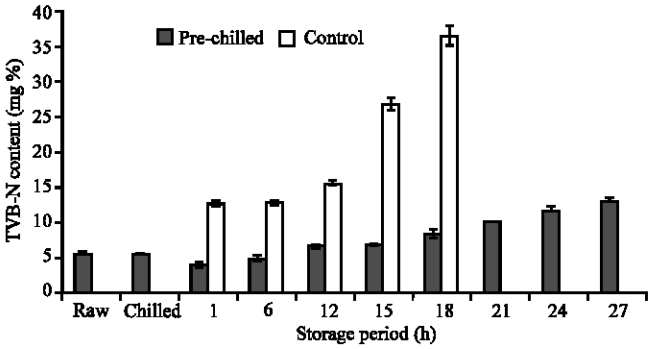

Fig. 7: Changes in TVB-N content of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

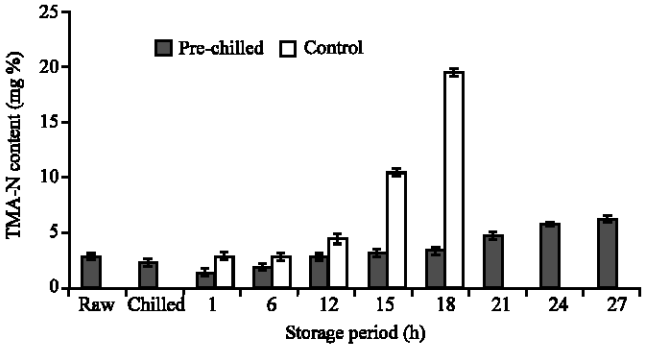

Fig. 8: Changes in TMA-N content of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

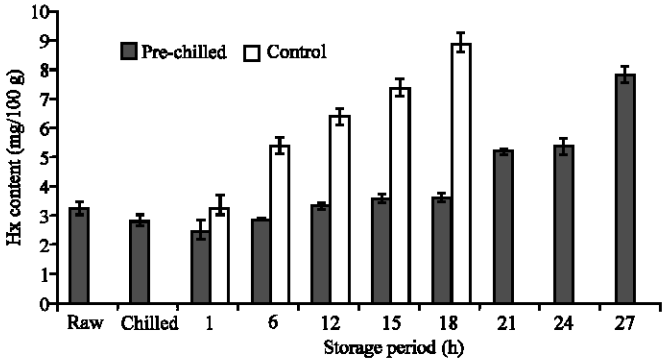

Fig. 9: Changes in hypoxanthine content of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

a combination of dry ice and ice at the ratio of 1:0.2:0.5 showed that hypoxanthine level reached to 8.20 mg/100 g at the end of storage period (Jeyasekaran *et al*., 2004a), which was almost similar to that of the present study.

Changes in Free Fatty Acid (FFA) content of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 10. Initial FFA content of fresh fish was 23.28% as oleic acid, which was in decreasing trend till the 12 and 6 h in pre-chilled and control pack, respectively. On the 15 and

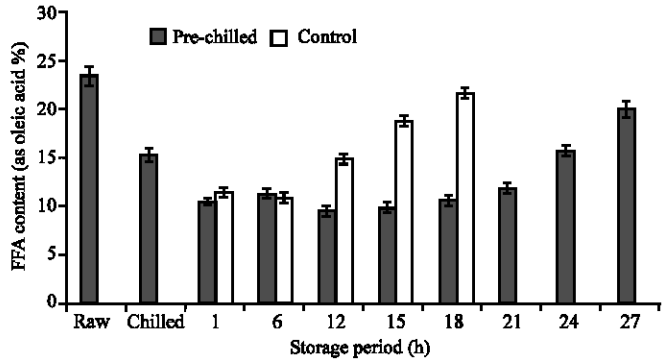

Fig. 10: Changes in free fatty acid content of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

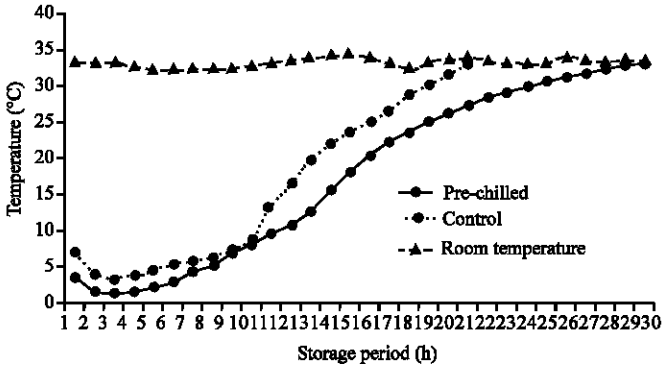

Fig. 11: Temperature profiles of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

12 h onwards, the content increased and reached to 19.84 and 21.58% as oleic acid in pre-chilled and control pack, respectively at the end of storage period and their changes were statistically significant ($p<0.05$). Jeyasekaran *et al*. (2004a) also observed a FFA level of 31.7% as oleic acid in dry ice and wet ice stored lethrinus (*Lethrinus miniatus*) at the end of storage period.

Temperature profiles of pre-chilled pomfret stored in a combination of dry ice and wet ice are given in Fig. 11. Temperature is one of the important factors influencing the growth of bacteria. After chilling with wet ice, immediately after packaging, the temperature recorded was 3.1°C. At that time, room temperature was 33.4°C. Low temperature of 1.3°C was observed at the 3 h of storage in pre-chilled pack and at that time control pack exhibited a temperature of 3.2°C. After that, temperature gradually increased in both the packages throughout the storage period. The results of present study confirmed the fact that the rate of deterioration spoilage is highly temperature dependent (Sivertsvik *et al*., 2002).

Changes in gas composition of pre-chilled pomfret stored in a combination of dry ice and wet ice are presented in Table 3. The atmospheric gas composition during this study was oxygen at 19.9%, carbon dioxide 0.5% and nitrogen 79.7%. On 1 h after packing, both the packages had a carbon dioxide level of 100%, which was due to the sublimation of dry ice inside the package. On further storage, the carbon dioxide level decreased slowly and simultaneously the oxygen and nitrogen levels increased due to the occupation of atmospheric air. Earlier studies reported that the longer shelf life obtained in dry ice packed fish might be due to high content of CO_2 in such packages (Dhananjaya and Stroud, 1994; Randell *et al*., 1999).

Table 3: Changes in gas composition of pre-chilled silver pomfret (*Pampus argenteus*) stored in a combination of dry ice and wet ice

Storage period (h)	Pre-chilled			Control		
	O_2 (%)	CO_2 (%)	N_2 (%)	O_2 (%)	CO_2 (%)	N_2 (%)
0	19.9	0.5	79.6	19.9	0.5	79.6
1	0.026	100.0	0.0	0.034	100.0	0.0
6	6.99	66.5	26.5	8.48	57.5	34.0
12	14.30	31.1	54.6	14.40	23.6	62.0
15	14.7	25.3	60.0	17.7	16.9	66.3
18	15.7	10.9	73.4	18.6	8.0	73.4
21	16.8	10.5	72.7	*DC	DC	DC
24	17.2	8.1	74.7			
27	18.6	7.8	73.6			

*DC-Discontinued

Conclusions

Based on the sensory, bacteriological, biochemical and physical quality analyses, the present investigation concluded that chilling before packaging in dry ice and wet ice extended the shelf life of silver pomfret (*Pampus argenteus*) by 50% and also resulted in better quality. Hence, the chilled fish exporters may adopt pre-chilling as a prerequisite for exporting fish in dry ice and wet ice to maintain the quality as well as increase the shelf life, in addition to considerable reduction in airfreight.

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