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Chilling Fresh Fish in Dry and Wet Ice

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Abstract

The use of dry ice combined with water ice for effective chilling of sardines *Sardinella gibbosa* was standardized by examining their sensorial, microbiological and biochemical qualities. Sardines stored in 50% dry ice alone had a longer shelf life of 10 days, when compared to that stored at 30 and 40% dry ice. The TPC, TMA-N, and TVB-N values were well within the level of acceptability up to 10 days. Sardines stored in 50% water ice along with the dry ice at the rate of 20 and 30% were found to be sensorially acceptable up to 13 days, and only 5 days with 10% dry ice. The TPC, TMA-N and TVB-N values of the sardines in the combination package with 20% dry ice and 50% water ice were also within the acceptable level.

Introduction

Fish, being highly perishable, spoil rapidly especially under tropical conditions. It is a known fact that fish that has been well handled and kept at low temperatures exhibit reduced bacterial growth. Dry ice as a cooling agent has certain advantages such as it removes three times the quantity of heat when compared to water ice, it has bacteriostatic effect and it acts as an insulant enveloping the fish upon evaporation (Putro 1989). Dry ice has been used for quite some time for the rapid transport of fresh fish by air, as it is weightless upon evaporation and does not pose leakage problems. The trend on the export of chilled fish from India is increasing as consumers in the international market prefer fresh fish than processed fish. Most of the exporters use only crushed water ice for chilling and transporting fresh fish at 100% level, which result to exorbitant transportation costs and leakage problems due to melting

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of ice. Application of dry ice is gaining popularity among fish traders involved in the short-term fresh fish transportation, as it considerably reduces the cost of transportation. A significant number of fish exporters use dry ice in combination with water ice at blind proportions for fresh fish transportation, as the use of dry ice alone is costly. However, there is no scientific report on the preservative effect of dry ice and its combination with water ice on the quality and shelf-life of fresh fishes. The present study was undertaken with the aim of standardizing the quantity of dry ice and its combination with water ice, required for effective chilling of fresh fish.

Materials and Methods

Materials

The fish sardines *Sardinella gibbosa* obtained from Tuticorin Fish Landing Centre, Tamil Nadu, India was selected as the raw material for this study. Dry ice (solid carbondioxide) was obtained from SIGGIL India Limited, Chennai. Flake ice produced by the flake ice maker (ZIEGRA-EIS-MASC HINEN, Germany) was used as the water ice. Thermocole boxes of 200 mm thickness lined with polythene sheets were used as the insulated container. Polythene bags were used for packing the water ice when it was used in combination with dry ice.

Trypticase Soya Agar supplied by Hi-Media, Mumbai, India was used for the microbiological analysis. The chemicals and reagents used for the analysis were of analytical grade or guaranteed reagent.

Experimental design

For the standardization of the quantity of dry ice, fresh sardines *Sardinella gibbosa* were eviscerated, washed and packed in thermocole boxes in three different levels of dry ice: 30%, 40% and 50%. Care was taken to avoid direct contact of fish with dry ice, by wrapping the ice in a brown sheet of paper and placing it at the four corners of the thermocole box. The boxes were closed air tight using cellophane tape and stored at room temperature. Dry ice loss due to evaporation was replenished by reicing every 24 h. Samples were drawn daily from each box for sensory, microbiological and biochemical analyses.

For the standardization of dry ice in combination with water ice, the sardines were also packed in thermocole boxes in three different levels of dry ice: 10%, 20% and 30%, along with 50% water ice. Water ice was packed separately in polythene bags and placed inside the thermocole boxes with the dry ice. Care was again taken to avoid direct contact of fish with dry ice. Similar procedure to that with dry ice pack was done. Reicing was done every 24 h to replenish lost dry ice as well as water ice due to evaporation and melting. Samples were likewise drawn every day for the sensorial, microbiological and biochemical changes.

Quality analysis

Sensory characteristics and overall acceptability of fish were assessed on the basis of 10 point scale described by Huss (1988). The total bacterial load was determined as per the methods described in APHA (1976). The TMA-N and TVB-N contents were estimated according to the method of Beatty and Gibbons (1937) using Micro conway diffusion units.

Statistical analysis

Analysis of variance (ANOVA) was performed (Snedecor and Cochran 1962) to examine whether any significant difference exists among the different packages of fish for the different quality characteristics.

Results and Discussion

Standardization of dry ice for effective chilling of fish

Figure 1 gives the sensorial quality changes of the sardines stored in different levels of dry ice. The over all sensory score of fish stored with 30% dry ice lowered drastically from 9.5 to 4.0 within four days of storage. But the sensory score of fish stored with 40 and 50% dry ice reached the acceptable limit of 4.0 on the 8^{th} and 10^{th} days of storage, respectively. At the highest concentration of dry ice (50%), the sardines exhibited longer shelf life, while at the lowest concentration (30%), it remained in acceptable condition only for four days. At the time of rejection the sardines gave off-odor, and exhibited flattened eyes and loss of scales. Connell (1995) reported that cod packed in 40%, 60% and 100% CO_2 gas and stored at 2ºC had an acceptable sensory score between the 9^{th} and 10^{th} days of storage; and there was no

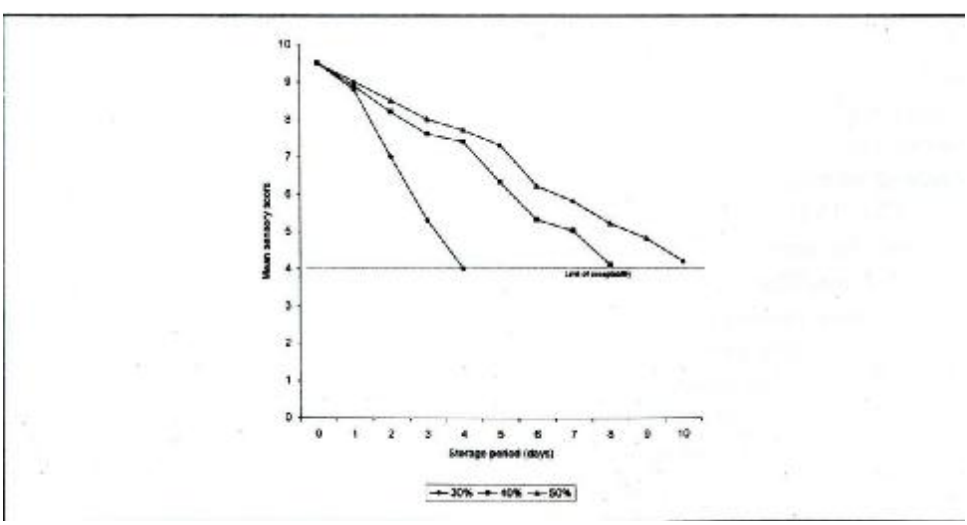

Fig. 1. Sensory quality of fish stored in different levels of dry ice.

remarkable difference on the shelf life at different concentrations of CO_2. However, in the present study significant differences ($p < 0.05$) were observed in the shelf life of sardines packed in three different concentrations of dry ice and at higher concentrations of dry ice. The effect is more pronounced, which is reflected in longer shelf life.

The total bacterial count of sardines stored in three different levels of dry ice are presented in table 1. The bacterial load increased from an initial load of 10^4 cfu·g^{-1} to a maximum of 10^7 cfu·g^{-1} during the storage period ($p < 0.05$). Fish stored in 30% dry ice reached a level of 10^7cfu·g^{-1} when they were found to be sensorially unacceptable. On the other hand, although the same level was attained in fish packed with 40% dry ice on the 6th day, they were found to be organoleptically acceptable. The bacterial load of fish stored with 50% dry ice did not reach the level of 10^7 cfu·g^{-1} even on the 10th day of storage. Callow (1932) reported that the displacement of O_2 by CO_2 inhibited the growth of aerobic microorganisms and resulted to a delayed growth phase. This implies that a higher level of CO_2 will displace more of the O_2 required for the microbial growth and prolong the lag phase of microbes, that had been reflected with an initial longer lag phase. The higher load of 10^7cfu·g^{-1} recorded in 30 and 40% dry ice stored sardines at the fag end of the storage might be due to the inadequacy of CO_2 to completely replace the O_2 needed for the microbial growth.

The changes in the TMA-N and TVB-N contents of sardines packed in different levels of dry ice are presented in figure 2. The TMA-N and TVB-N contents gradually increased ($p < 0.05$) in all the packages of dry ice upon storage. The TMA-N level of fish stored with 30% dry ice increased to a maximum of 8.18 mg% on the 4th day of storage, whereas it reached an almost similar level in the fish stored with 40% dry ice only on the 8th day of storage. On the other hand, the TMA-N level was much less in the fish stored with 50% dry ice and could reach only up to 4.79 mg% at the end of the storage period. The TMA-N content in 40 and 50% dry ice packed sardines was found to remain more or less constant for nearly three days and thereafter a gradual increase was observed. The TMA-N contents in all the packages of sardines were within the acceptable limit of 10 to 15 mg% (Connell 1995), although they were organoleptically rated as unacceptable. The results confirm the findings of Oberlender et al. (1983) who reported that the TMA-N content of CO_2 stored sword fish steaks did not exceed the acceptable level of freshness even after 20 days of storage at 20^{o}C.

The TVB-N contents of sardines stored in 30 and 50% dry ice did not exceed the acceptable limit of 30 mg% (Connell 1995) in sardines stored in 40% dry ice. The level slightly exceeded this limit (Fig. 2) at the time of rejection. A very gradual increase in the TVB-N content was observed in sardines stored with 50% dry ice from the 1st day to the 7th day of storage (12 to 16 mg%) which later increased progressively. The lower TMA-N and TVB-N contents observed in the 50% dry ice packed sardines when compared to other packages is mainly because of the effect of higher quantity of CO_2 in inhibiting the microorganisms responsible for the formation of TMA-N and TVB-N in fish muscle (Oberlender et al. 1983).

Table 1. Changes in total bacterial load (cfu·g^{-1}) in fish stored in different levels of dry ice.

Storage period (days)	30%	40%	50%
0	2.10 x 10^4	2.10 x 10^4	2.10 x 10^4
1	4.80 x 10^5	1.30 x 10^5	3.30 x 10^4
2	1.96 x 10^6	6.00 x 10^5	7.00 x 10^4
3	2.18 x 10^6	7.80 x 10^5	5.30 x 10^4
4	1.02 x 10^7	9.50 x 10^5	2.24 x 10^5
5	DC	8.60 x 10^6	4.30 x 10^5
6		5.40 x 10^7	5.80 x 10^5
7		6.10 x 10^7	7.80 x 10^5
8		8.40 x 10^7	9.20 x 10^5
9		DC	1.20 x 10^6
10			3.96 x 10^6
11			DC

DC- Discontinued

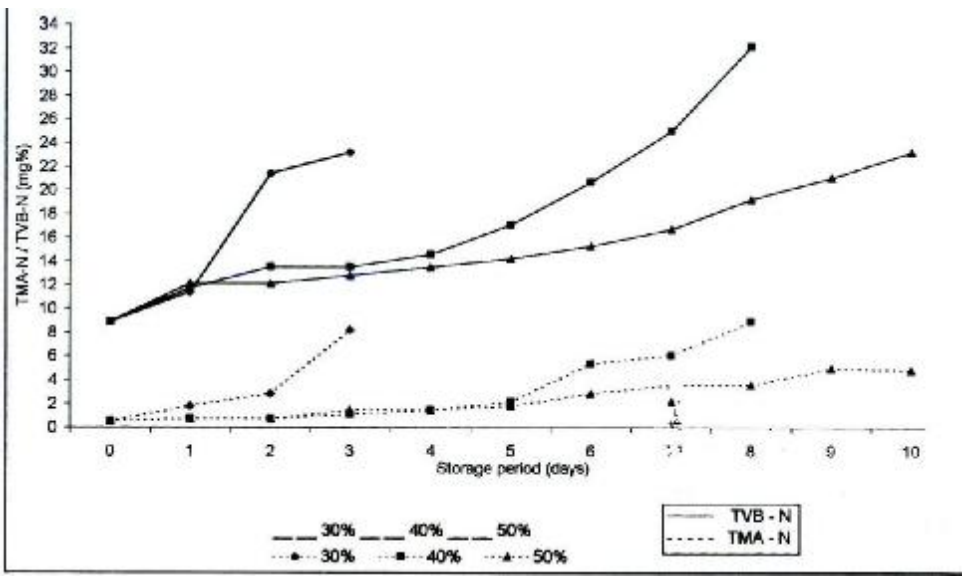

Fig. 2. Changes in trimethyl amine nitrogen (TMA-N) / total volatile base-nitrogen (TVB-N) content (mg%) of fish stored in different levels of dry ice.

Standardization of dry ice in combination with water ice for effective chilling of fish

Water ice is commonly used for preserving fish at a ratio of 1:1. To reduce the cost of transportation, instead of using 100% water ice, the quantity was brought down to 50% and kept constant. Figure 3 shows the sensorial quality of the sardines stored in three different levels of dry ice: 10%, 20% and 30% along with 50% water ice. The sardines stored in 10% dry ice became sensorially unacceptable within six days, due to the loss of characteristics appearance, odor and texture. But the packages of sardines stored in 20% and 30% dry ice exhibited more or less sensorial quality with a shelf life of 13

days. Putro (1989) had observed that incorporation of dry ice improved the efficiency of water ice used for chilling fresh fish, particularly during bulk handling. A similar effect was also noticed in the present study. Dry ice even at 20% level could extend the shelf life of sardines up to 13 days when packed with 50% water ice.

Changes in the total bacterial load of the sardines packed in different levels of dry ice along with 50% water ice are presented in table 2. The bacterial load of the sardines stored in 10% dry ice exceeded the permissible limit of 10^6cfu·g^{-1} (Shewan 1977) within three days of storage, whereas in sardines stored in the other two packages, the same level was attained only after 12 days of storage. The delayed microbial growth in the latter packages was mainly due to the higher concentration of dry ice. No significant difference ($p > 0.05$) was observed in the microbial quality of the sardines stored in 20 and 30% dry ice along with 50% water ice. It is believed that in these

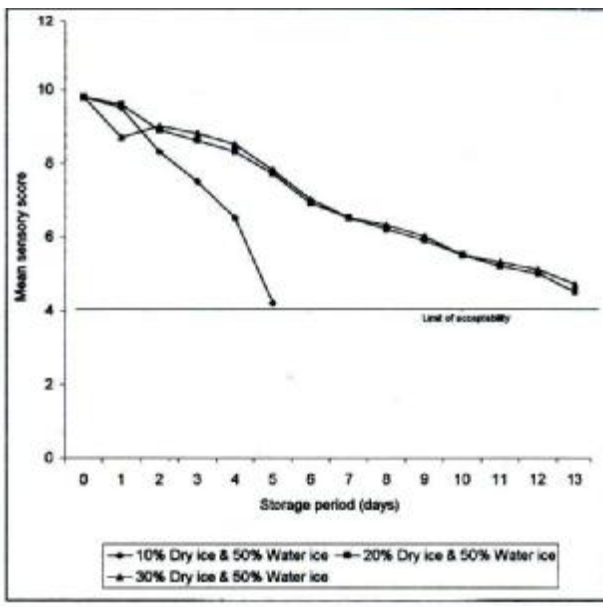

Fig. 3. Sensory quality of fish stored in different combinations of dry ice with water ice.

Table 2. Changes in total bacterial load (cfu·g^{-1}) of fish stored in different combinations of dry ice and water ice.

Storage period (days)	10%* & 50%**	20%* & 50%**	30%* & 50%**
0	3.80×10^4	3.80×10^4	3.80×10^4
1	2.20×10^5	4.90×10^4	4.33×10^4
2	4.00×10^6	5.65×10^4	5.61×10^4
3	1.96×10^7	6.25×10^4	6.40×10^4
4	2.26×10^7	7.80×10^4	7.10×10^4
5	5.60×10^7	9.70×10^4	9.65×10^4
6	DC	1.07×10^5	1.04×10^5
7		3.30×10^5	2.85×10^5
8		8.60×10^5	8.25×10^5
9		1.50×10^6	1.30×10^6
10		4.20×10^6	4.00×10^6
11		5.53×10^6	5.27×10^6
12		6.40×10^6	2.66×10^6
13		1.72×10^6	1.58×10^7
14		DC	DC

DC - Discontinued
*% of dry ice
**% of water ice

packages, the multiplication of microbes was initially suppressed (i.e. up to five days) due to the chilling effect of the higher concentration of dry ice. Huss (1988) reported that in tropical fish chilled in water ice immediately after capture, there was an initial reduction in the microbial load, which later increased upon storage. The same effect was noticed in the present study even in fish chilled with dry ice and water ice.

Figure 4 depicts the changes in the TMA-N and TVB-N of sardines stored in 50% water ice along with different levels of dry ice along with water ice. Initially, the TMA-N content of sardines was very low and gradually increased ($p < 0.05$) upon storage. The TMA-N content did not exceed the limit of acceptability (Connell 1995) in all the packages of sardines even at the time of rejection. A similar result was also observed in the case of TVB-N content of sardines with the values below the maximum limit of acceptability of 30 mg% (Connell 1995). The formation of TMA-N and TVB-N was comparatively less in the 20 and 30% dry ice stored sardines than in the 10% dry ice package as the dry ice at 20% concentration or more could lower the storage temperature and delay the accumulation of total volatile base nitrogen compounds in fish.

The TMA-N and the TVB-N contents were found to be unreliable indicators of quality as the sardines became organoleptically unacceptable even as the TMA-N and the TVB-N values were within the maximum limit of acceptability.

Comparison of the effect of dry ice and its combination with water ice

From the above observation, it was found that among the sardines stored in different levels of dry ice, 50% dry ice packed sardines exhibited a maximum shelf life of 10 days with an acceptable microbiological and biological

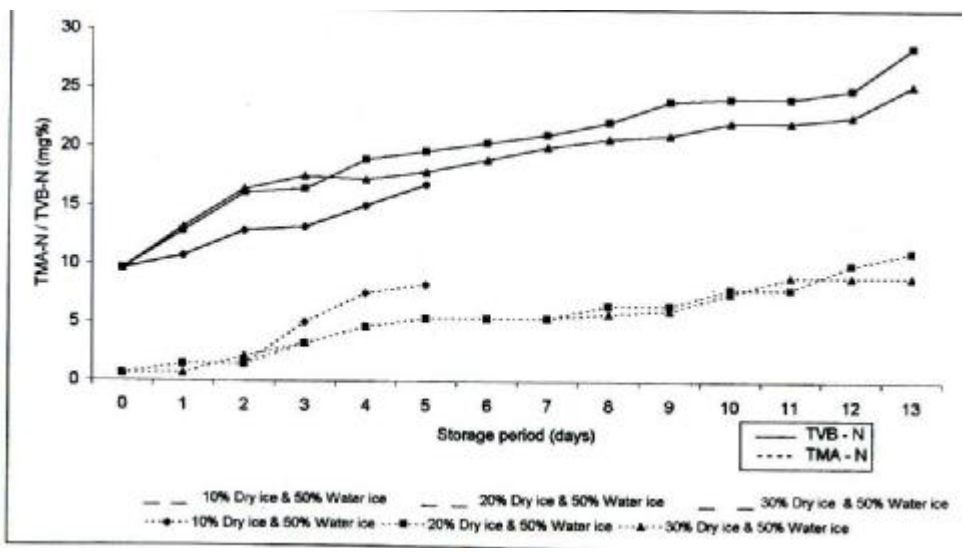

Fig. 4. Changes in trimethyl amine-nitrogen (TMA-N) / total volatile base-nitrogen (TVB-N) content (mg%) of fish stored in different combinations of dry ice with water ice.

quality. On the other hand, the sardines packed in 20 and 30% dry ice along with 50% water ice, yielded a better quality product with a shelf life of 13 days. As the 20 and 30% concentrations of dry ice resulted in similar shelf life in the combination package, the lower level of 20% seemed to be sufficient for effective packaging as it is economical. The results therefore clearly show that the shelf life of the sardines can be extended by three days when packed in combination of dry ice and water ice. Taking into consideration the overall quality, the sardines packed in combination of dry ice and water ice were found to be superior than that packed with dry ice alone.

Conclusion

The results suggest that 50% dry ice is necessary to come up with a better quality product with a reasonable shelf life (10 days), when sardines are packed in dry ice alone. When they are packed with the combination of dry ice and water ice, 20% dry ice and 50% water ice is sufficient to yield a good quality product with better shelf life (13 days).

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