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# The effect of different precooling media during processing and cooling techniques during packaging of cod (*Gadus morhua*) fillets

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## Report summary

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<i>Ágríp á íslensku:</i>	<p>Tilgangur tilraunanna var að kanna áhrif mismunandi kælimiðla við forkælingu fyrir pökkun á hitastýringu, gæði og geymsluþol þorskflaka. Eftirfarandi kælimiðlar voru kannaðir og bornir saman við enga sérstaka forkælingu fyrir pökkun:</p> <p>1) þekill með lágu saltinnihaldi, 2) krapaís með lágu saltinnihaldi.</p> <p>Auk þess voru könnuð áhrif þess að nota annars vegar ísmottur og hins vegar þurrís við geymslu flakanna. Fylgst var með breytingum á hitastigi með hitanemum á öllum stigum. Sýni voru gæðametin með skynmati, örveru- og efnamælingum í allt að 13 daga frá vinnslu og pökkun (16 daga frá veiði). Flökin voru geymd við ofurkældar aðstæður (undir 0 °C) mestan hluta geymslutímans.</p> <p>Lægra hitastig krapaíss en þekills leiddi til lægra hitastigs flaka við pökkun auk þess sem hiti vökvapækilsins reyndist hækka hratt þegar hlé var gert á vinnslu.</p> <p>Mismunandi meðhöndlun leiddi til sambærilegs ferskleikatíma samkvæmt skynmati. Hins vegar reyndist notkun vökvapækils við forkælingu fyrir pökkun leiða til 1 – 2 daga skemmra geymsluþols samanborið við enga forkælingu eða forkælingu með krapaís. Rekja má ástæður þessa til þess að þekillinn innihélt töluvert magn örvera, m.a. H<sub>2</sub>S-myndandi gerla sem eru virkir framleiðendur á trímetylamíni (TMA). Samanburður á vökvakældu flökunum sýndi að notkun á þurrís lengdi geymsluþol um 1 dag í samanburði við ísmottur. Niðurstöður örveru- og efnamælinga voru í samræmi við þessar niðurstöður.</p>		
<i>Lykilorð á íslensku:</i>	<i>Forkæling, kælimiðlar, þorskflök, ferskleiki, geymsluþol</i>		

*Summary in English:*

The aim of the experiment was to investigate effects of two cooling media during precooling at processing on temperature control, quality and storage life of cod fillets. The two cooling media compared to no special precooling during processing (NC) were: 1) liquid brine (LC) and 2) slurry ice (SIC). In addition, the influence of using either dry ice or ice packs during storage was studied. The samples were kept at superchilled conditions during most of the trial. The environmental and product temperature history of each group was studied using temperature monitors. The samples were analysed with sensory evaluation, microbial and chemical methods for up to sixteen days from catch (thirteen days from processing).

Lower temperature of the slurry ice than the liquid brine resulted in lower fillet temperature at packaging and the liquid brine temperature increased rapidly during a processing break, which seems to be a weakness of the liquid brine tank.

Results from sensory, microbial and chemical analysis all showed that immersing the skinless cod fillets in liquid cooling brine prior to packaging resulted in one to two days reduction of shelf life in comparison with fillets that were not immersed in liquid brine (no cooling) or in slurry ice. This could be attributed to the fact that the cooling brine carried considerable amounts of microbes including H<sub>2</sub>S-producing bacteria which are active producers of trimethylamine (TMA). Comparison of the groups receiving liquid cooling showed that dry ice appeared to extend the shelf life of one day as compared to ice packs. The length of the freshness period was, however, similar in all experimental groups according to sensory evaluation. These results were confirmed by total volatile bases (TVB-N) and TMA analysis and microbial counts.

*English keywords:*      *Precooling, cooling media, cod fillets, freshness, shelf life*

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# 1 INTRODUCTION

Various precooling methods are applied during processing and packaging of fresh fish fillets in order to lower the product temperature and thereby protect it against thermal abuse during transport and prolong shelf life. Different types of slurry ice mixtures and liquid brines are among the utilised precooling media and different types of phase change materials (cooling packs containing gel or frozen water ice) and dry ice are applied during packaging of the fresh fish fillets.

The aim of the experiment was to investigate effects of two cooling media during precooling at processing on the storage life of cod fillets. The two cooling media compared to applying no special precooling during processing (NC) were: 1) liquid brine (LC) and 2) slurry ice (SIC). In addition, the influence of using either dry ice or ice packs during storage was studied. The samples were kept at superchilled conditions during most of the trial. The environmental and product temperature history of each group was studied using temperature monitors. The samples were analysed with sensory evaluation, microbial and chemical methods for up to 16 days from catch (13 days from processing).

## 2 MATERIAL AND METHODS

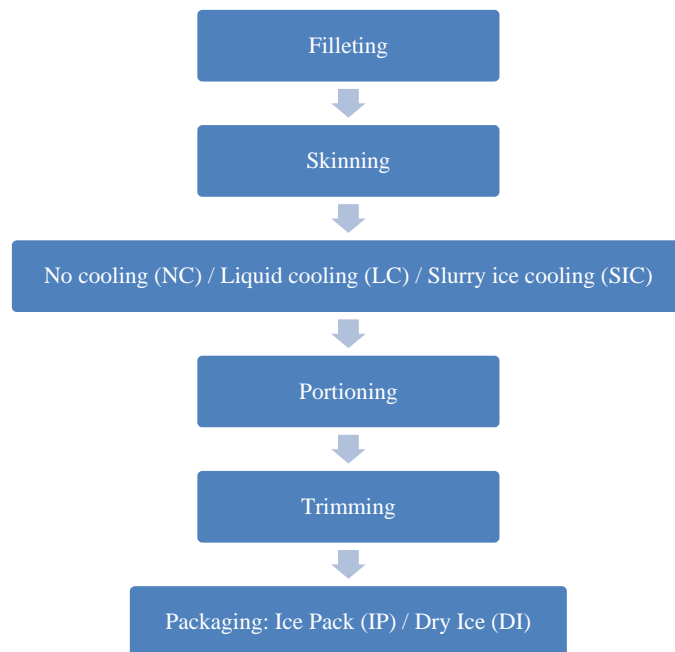
### 2.1 Experimental design

Cod used in the experiment was caught by a trawler north of Iceland on Feb 13<sup>th</sup> 2010.



**Figure 1.** The trawler, Sólbakur EA 1, that caught the fish processed for the trial. ([http://www.ua.is/skipin/solbakur\\_ea\\_1/?nocache=true](http://www.ua.is/skipin/solbakur_ea_1/?nocache=true)).

After bleeding and gutting the cod was washed in sea-water on deck and then transported to the hold where it was iced with slurry ice in tubs. The cod was processed at a fish processing plant in North Iceland, Feb 16<sup>th</sup> 2010. The fish was filleted and fillets without skin got different treatment (see below) prior to packaging in 5 kg expanded polystyrene (EPS) boxes. After packaging the cod was transported to Matís ohf. where it was stored at superchilled temperature conditions inside an air climate chamber for up to 13 days from catch (16 days from catch).



**Figure 2: Different cooling and packaging methods during processing of cod at Brim fish processing plant**

The experimental groups were as follows:

1. NC-IP: No cooling after skinning, ice pack (250 g) applied during packaging.
2. LC-IP: Liquid cooling after skinning, ice pack applied during packaging.
3. LC-DI: Liquid cooling after skinning, dry ice applied during packaging.
4. SIC-DI: Slurry ice cooling (see Figure 3) after skinning, dry ice applied during packaging (see Figure 4).



**Figure 3. Implementation of the slurry ice cooling (SIC) after skinning.**



**Figure 4. Dry ice (DI) on top of fillets in an EPS box.**

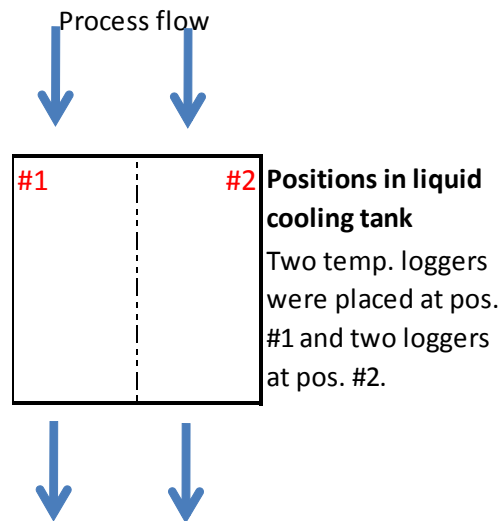
## **2.2 Cooling medium sampling during processing**

Samples for microbial, pH and salinity analysis were taken from the liquid cooling tank at 07:30, 08:25, 09:20, 10:30 and 10:50. Each time one sample was taken from the front end of the tank and another sample from the back end of the tank. One sample for salinity analysis was taken from the slurry ice at 09:25.

## **2.3 Temperature measurements**

Micro-T DS1922L temperature loggers from NexSens Technology (Dayton, OH, USA, see Figure 6) were used for all temperature monitoring in the trial. This logger has an

accuracy of  $\pm 0.5$  °C and a resolution of 0.0625 °C and an operating range of -40 to 85 °C. The diameter is 17 mm and the thickness is 5 mm. The cooling medium temperature was mapped both during the liquid cooling (see Figure 5) and slurry ice cooling during processing.



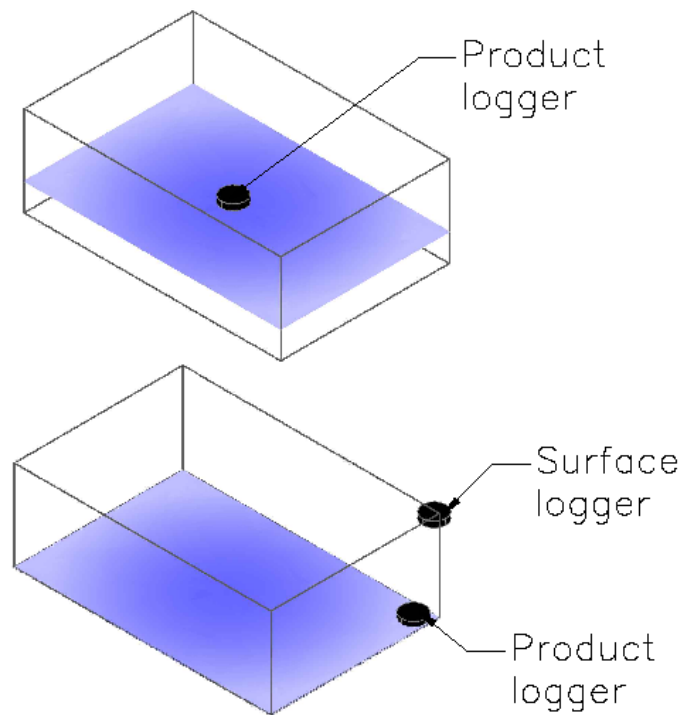
**Figure 5. Relative positions of temperature loggers in the liquid cooling tank.**

After packaging, product and surface temperatures were monitored for two EPS boxes in each of the four experimental groups. Two Micro-T temperature loggers were put in each of the monitored EPS boxes and one on the outside of each box, see Figure 7. For measuring the product temperature, the loggers were placed in plastic bags, in order to avoid microbial contamination. Temperature was monitored at two positions on the air climate chamber floor at Matís in addition to one position at the chamber wall.



**Figure 6. Micro-T DS1922L temperature loggers.**





**Figure 7. Positions of surface temperature logger and product temperature loggers at mid-height (above) and bottom (below) of EPS boxes.**

## **2.4 Sensory evaluation**

Quantitative Descriptive Analysis (QDA), introduced by Stone and Sidel (2004), and the Torry freshness score sheet (Shewan and others 1953) were used to assess cooked samples of cod. Nine panellists all trained according to international standards (ISO 1993); including detection and recognition of tastes and odours, trained in the use of scales and in the development and use of descriptors participated in the sensory evaluation. The members of the panel were familiar and experienced in using the QDA method and Torry freshness score sheet for cod. The panel was trained in recognition of sensory characteristics of the samples and describing the intensity of each attribute for a given sample using an unstructured scale (from 0 to 100%). Most of the attributes were defined and described by the sensory panel during other projects (Sveinsdottir and others 2009). The sensory attributes were 30 and are described in Table 2.

Samples weighing ca. 40 g were taken from the loin part of the fillets and placed in aluminium boxes coded with three-digit random numbers. The samples were cooked to a core temperature of 67 °C in a pre-warmed oven (Convotherm Elektrogeräte GmbH, Eglfing, Germany) at 95-100 °C with air circulation and steam, and then served to the panel. Each panellist evaluated duplicates of each sample in a random order in ten sessions (maximum four samples per session).

A computerized system (FIZZ, Version 2.0, 1994-2000, Biosystèmes) was used for data recording.

**Table 1: Sensory vocabulary for cooked samples of cod (*Gadus morhua*)**

Sensory attribute	Short name	Description of attribute
<b>Odour</b>		
sweet	o-sweet	sweet odour
shellfish, algae	o-shellfish	shellfish, algae, characterict fresh odour
meaty	o-meat	meaty odour, reminds of boiled meat or halibut
vanilla, boiled milk	o-vanilla	vanilla, sweet boiled milk
boiled potatoes	o-potatoes	odour reminds of whole, warm, boiled potatoes
frozen storage	o-frozen	reminds of odour found in refrigerator and/or freezing compartment
table cloth	o-cloth	reminds of damp, unclean cloth (left on kitchen table for 36 h)
TMA	o-TMA	TMA odour, reminds of dried salted fish, amine
sour	o-sour	sour odour, spoilage sour, acetic acid
sulphur	o-sulphur	sulphur, matchstick, boiled kale
<b>Appearance</b>		
light/dark colour	a-dark	Left end: light, white colour. Right end: dark, yellowish, brownish, grey
homogenous/ heterogeneous	a-heterog.	Left end: homogenous, even colour. Right end: discoloured, heterogeneous, stains
white precipitation	a-prec.	white precipitation in the broth or on the fish
<b>Flavour</b>		
salt	f-salt	salt taste
metallic	f-metallic	metallic flavour
sweet	f-sweet	characteristic sweet flavour of very fresh (boiled) cod
meaty	f-meat	meaty flavour, reminds of boiled meat
frozen storage	f-frozen	reminds of food which has soaked in refrigerator/freezing odour
pungent	f-pungent	pungent flavour, bitter
sour taste	f-sour	sour taste, spoilage sour
TMA	f-TMA	TMA flavour, reminds of dried salted fish, amine
off flavour	f-off	strenght of off flavour (spoilage flavour/off-flavour)
<b>Texture</b>		
flakiness	t-flakes	the fish portion slides into flakes when pressed with the fork
firm/soft	t-soft	Left end: firm. Right end: soft. Evaluate how firm or soft the fish is during the first bite
dry/juicy	t-juicy	Left end: dry. Right end: Juicy. Evaluated after chewing several times: dry - pulls juice from the mouth
tough/tender	t-tender	Left end: tough. Right end: tender. Evaluated after chewing several times
mushy	t-mushy	mushy texture
meaty	t-meaty	meaty texture, meaty mouth feel, grude muscle fibers
clammy	t-clammy	clammy texture, dry red wine, tannin
rubbery	t-rubbery	rubbery texture, springy

## 2.5 Microbial measurements

Total viable psychrotrophic counts (TVC) and counts of H<sub>2</sub>S-producing bacteria were evaluated on iron agar (IA) as described by Gram and others (1987) with the exception that 1% NaCl was used instead of 0.5% with no overlay. Plates were incubated at 17 °C for 4-5 d. Bacteria forming black colonies on IA produce H<sub>2</sub>S from sodium thiosulphate and/or cysteine. Cephaloridine Fucidin Ceftrimide (CFC) agar was modified according to Stanbridge and Board (1994) and used for enumeration of presumptive pseudomonads. Pseudomonas Agar Base (Oxoid) with CFC Selective Agar Supplement (Oxoid) was used. Plates were incubated at 22 °C for 3 d. *Pseudomonas* spp. form pink colonies on this medium. In all the above counts surface-plating was used.

Counts of *Photobacterium phosphoreum* were obtained by a quantitative PCR method. Briefly, One ml of the tenfold diluted fish samples in Maximum Recovery Diluent (MRD, Oxoid) buffer was frozen at -20 °C for later DNA extraction. For the DNA extraction, the diluted samples were centrifuged at 11.000 x g for 7 min to form a pellet. The supernatant was discarded and DNA was recovered from the pellet using the Promega Magensil KF, Genomic system (MD1460) DNA isolation kit (Promega Corporation, Madison, USA) in combination with KingFisher magnetic beads automatic DNA isolation instrument (Thermo LabSystems, Waltham, USA) according to the manufacturers' recommendations. All PCR reactions were done using the Mx3005p instrument. The PCR was done using Brilliant QPCR mastermix (Stratagene, La Jolla, CA, USA). Primers were synthesized and purified with HPLC (MWG, Ebersberg, Germany). The DNA standard used for quantification was previously calibrated against the PPDM-Malthus conductance method (Dalgaard and others 1996; Lauzon 2003). Cooled MRD buffer was used for all dilutions. All samples were analysed in duplicate and results presented as an average.

## 2.6 Chemical analysis

### 2.6.1 Total Volatile Base Nitrogen and Trimethylamine

The method of Malle and Tao (1987) was used for total volatile bases (TVB-N) and trimethylamine (TMA) measurements. TVB-N was measured by steam distillation (Struer TVN distillatory, STRUERS, Copenhagen) and titration, after extracting the fish

muscle with 7.5% aqueous trichloroacetic acid solution. The distilled TVB-N was collected in boric acid solution and then titrated with sulphuric acid solution. TMA was measured in trichloroacetic acid (TCA) extract by adding 20 ml of 35% formaldehyde, an alkaline binding mono- and diamine, TMA being the only volatile and measurable amine. All chemical analyses were done in duplicate.

### 2.6.2 *pH - measurements*

The pH was measured in 5 grams of minced loins mixed with 5 mL of deionised water using the Radiometer PHM 80. The pH meter was calibrated using the buffer solutions of pH  $7.00 \pm 0.01$  and  $4.01 \pm 0.01$  (25 °C) (Radiometer Analytical A/S, Bagsvaerd, Denmark).

### 2.6.3 *Salt and water content*

The water content of each fillet was measured by accurately weighing out 5 grams of the minced sample in a ceramic bowl with sand. The sample was then mixed to the sand and dried in an oven at  $103 \pm 2$  °C for 4 hours. The water content was based on weight differences before and after the drying of three replicates for each sample (ISO 6496, 1999). Salt content was measured with the Volhard Titrimetric method according to AOAC ed. 17 from 2000 (no. 976.18).

## 2.7 **Water holding capacity and drip**

The water holding capacity (WHC) was determined by a centrifugation method (Eide and others 1982). Approximately 2 g of the minced fish was weighed accurately and centrifuged (Heraeus Biofuge Stratos, Kendro Laboratory products, USA) at  $210 \times g$ , for 5 minutes at 0-5°C. The weight lost during centrifugation ( $\Delta m_{centrifuged}$ ) was evaluated as water loss and no corrections made for other components as may be necessary for fishes with high fat content. WHC was calculated as the ratio of the water retained in the sample, compared to mass of water before centrifugation ( $m_t * x_t^w$ ):

$$WHC = \left( \frac{m_t * x_t^w - \Delta m_{centrifuged}}{m_t * x_t^w} \right) * 100$$

Drip was evaluated fourteen days after processing, i.e. one day after the last sampling day (d13). It was evaluated by measuring the weight of the liquid which drained from the fillets in the boxes. The weight of the fish was also recorded. The drip was then calculated as the ratio of the liquid lost during storage to the original weight of the fish.

## 2.8 Data analysis

Principal Component Analysis (PCA) on significant mean values of QDA sensory attributes was performed, using full cross validation. Analysis of variance (ANOVA) was carried out on QDA and Torry data in the statistical program NCSS 2000 (NCSS, Utah, USA). The program calculates multiple comparisons using Duncan's multiple comparison test. The significance level was set at 5%, if not stated elsewhere.

### 3 RESULTS AND DISCUSSION

#### 3.1 Temperature measurements

##### 3.1.1 *Temperature in liquid cooling medium*

The temperature evolution at two different positions in the liquid cooling tank is shown in Figure 8, see relative positions in the tank in Figure 5. It should be noted that the experimental groups LC-IP and LC-DI were collected between 07:45 and 08:15 AM when the temperature of the liquid was between -0.4 and 0.6 °C. The liquid temperature in the liquid cooling tank is similar to what Margeirsson and Arason (2009) measured in February 2009 but 1 – 2 °C lower than Magnusson and others (2009) and Valtýsdóttir and others (2009) found in Feb. 2009 and June 2009, respectively. The sojourn time in the liquid cooling tank was 6 - 7 minutes. The results presented in Figure 8 are only relevant for these two groups (LC-IP and LC-DI).

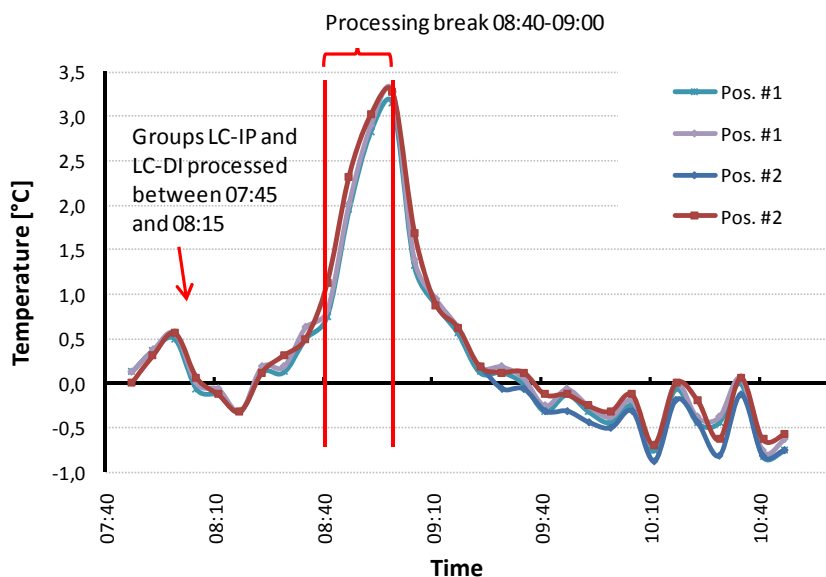
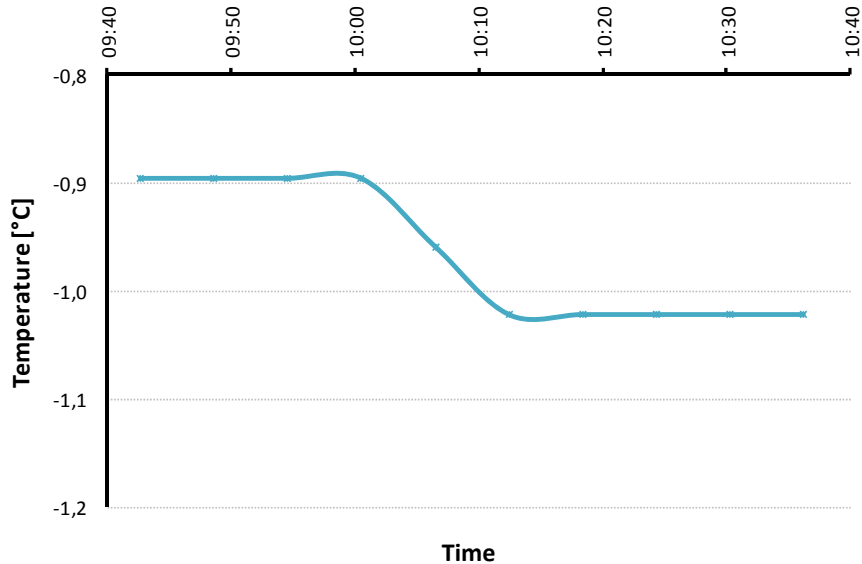


Figure 8. Temperature in the liquid cooling tank applied for experimental groups LC-IP and LC-DI.

##### 3.1.2 *Temperature in slurry ice cooling medium*

The temperature evolution of the slurry ice in the slurry ice cooling tub during the period which group SI-DI was processed is shown in Figure 9. The results presented in Figure 9 are only relevant for the experimental group SIC-DI and the sojourn time of fillets in the slurry ice tub was 6 – 7 minutes as for the LC-groups. By comparing Figure 9 to Figure 8

it is noted that significantly lower cooling medium temperature was experienced for slurry ice than the liquid in the liquid cooling tank.



**Figure 9.** Temperature of the slurry ice applied for experimental group SIC-DI.

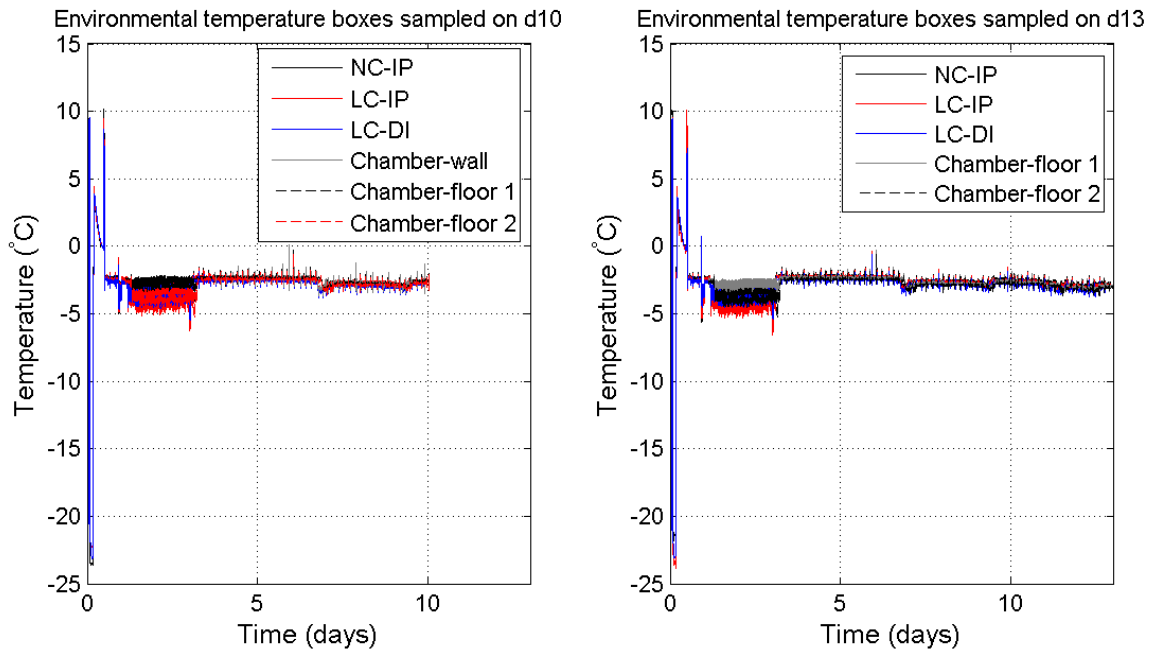
### 3.1.3 *Environmental temperature after packaging*

The environmental (surface and ambient air) temperature evolution is shown in Figure 10 and the mean surface temperature during the whole experiment is presented in Table 2. No results are presented for the group SIC-DI since the surface temperature logger failed, however, judging from the other positions, the environmental temperature distribution was very homogenous throughout the trial. The environmental temperature inside the air climate chamber was measured as  $(-2.7 \pm 0.3)$  °C both at the floor and at the chamber wall at 1.0 m height.

**Table 2.** Mean and standard deviations of surface temperatures for boxes sampled on day 13 (d13).

Group	Mean (°C)	Standard deviation (°C)
NC-IP	-2.6	2.1
LC-IP	-2.9	2.3
LC-DI	-3.0	2.3





**Figure 10. Environmental temperature after packaging at the processing plant during land transport to Matis, where it was stored in an air climate chamber for 13 days. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.**

### 3.1.4 *Product temperature*

The product temperature evolution is shown in Figure 11 and the mean product temperature during the whole experiment is presented in Table 3. Mean and standard deviations of product temperatures for boxes sampled on day 10 (d10).

Group	Mean (°C)	Standard deviation (°C)
NC-IP	-1.9	0.9
LC-IP	-1.0	0.4
LC-DI	-1.1	0.1
SIC-DI	-1.4	0.1

Table 4 and Table 3. The mean and standard deviations of the product temperatures in the tables are calculated from the mean of the temperatures at the two positions in each box. The product temperature of the NC-IP group was interestingly lower than for the other groups, this can be seen both in the boxes sampled on d10 and d13. The rapid temperature increase shown after around seven days inside the d13-NC-IP box (Figure 11-right) could possibly be explained with failure of both product temperature loggers in the d13-NC-IP box at least when compared to the results for d10-NC-IP (Figure 11-left). The mean product temperature in the d10-NC-IP box was  $(-1.9 \pm 0.9)$  °C, i.e.

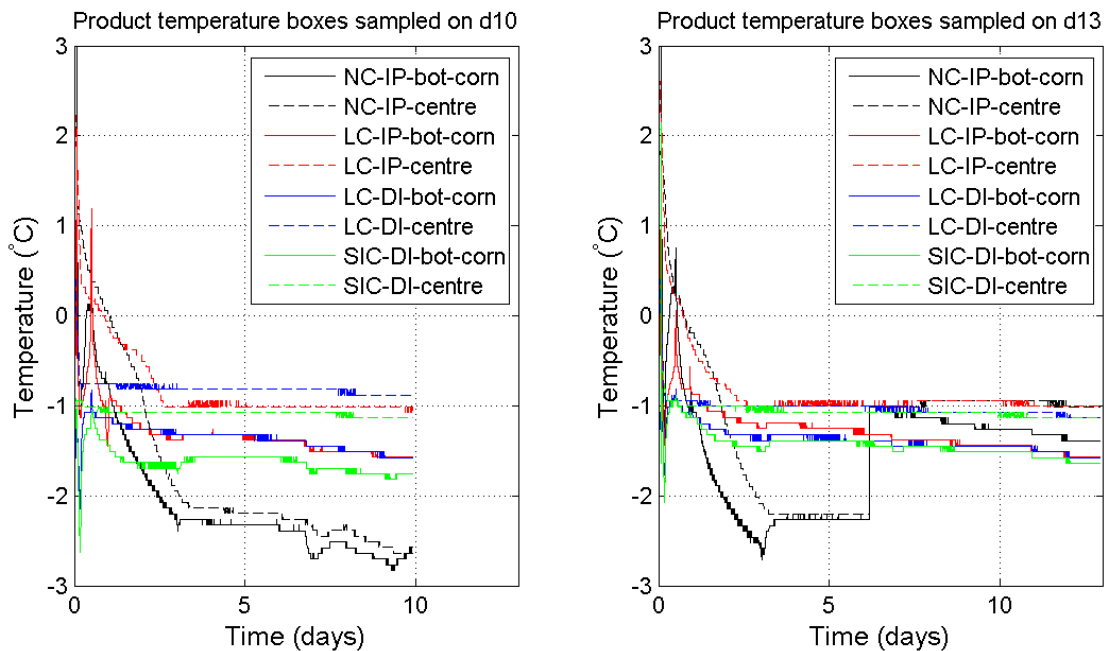
significantly lower than in the corresponding d13-NC-IP box and actually, lower than in all other boxes in the trial.

**Table 3. Mean and standard deviations of product temperatures for boxes sampled on day 10 (d10).**

Group	Mean (°C)	Standard deviation (°C)
NC-IP	-1.9	0.9
LC-IP	-1.0	0.4
LC-DI	-1.1	0.1
SIC-DI	-1.4	0.1

**Table 4. Mean and standard deviations of product temperatures for boxes sampled on day 13 (d13).**

Group	Mean (°C)	Standard deviation (°C)
NC-IP	-1.4	0.7
LC-IP	-1.1	0.3
LC-DI	-1.2	0.1
SIC-DI	-1.2	0.2

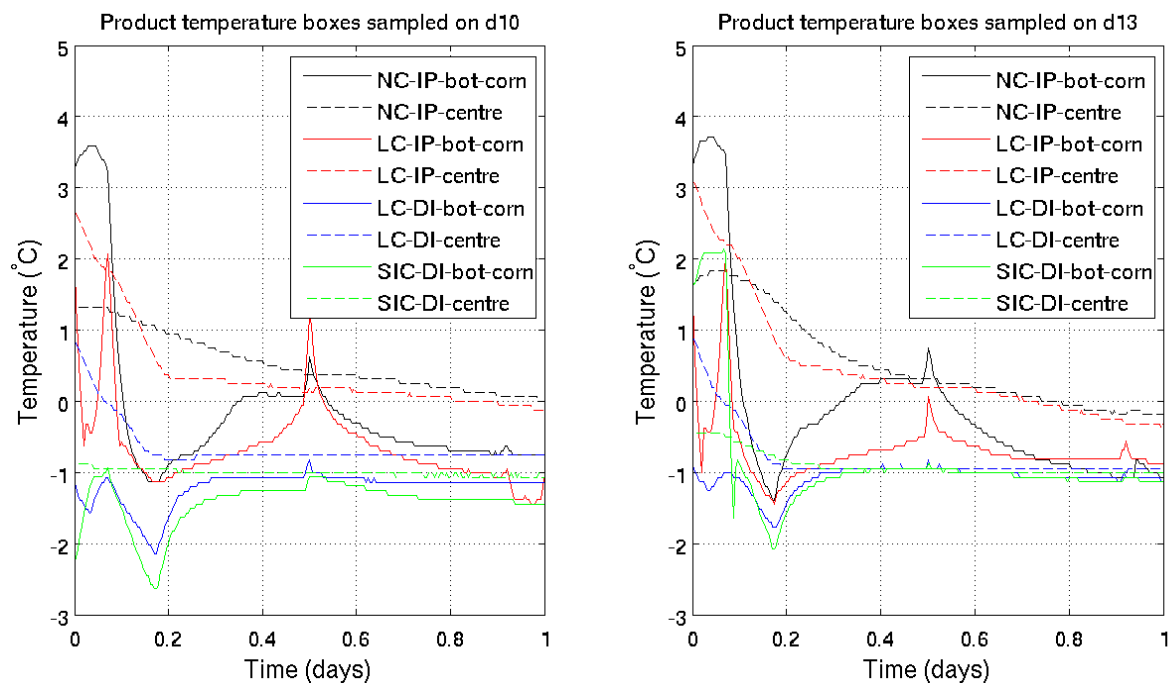


**Figure 11. Product temperature after packaging at the processing plant during land transport to Matis, where it was stored in an air climate chamber for 13 days. Left: boxes sampled on day 10.**

**Right: boxes sampled on day 13. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.**

Furthermore, Figure 11 shows that the bottom corner location is more sensitive to external temperature load (which was negative for most of the time, i.e. environmental temperature was lower than product temperature), where it responds much faster to environmental temperature changes than the temperature at the centre of the box. At the centre of the box the temperature response is dampened relying on the thermal capacity of the product itself. For all other groups than the NC-IP group, the temperature was around 0.5 – 0.8 °C lower at the centre than at the bottom corner of the investigated boxes.

Figure 12 presents a zoom up of the product temperature in Figure 11 during the first day of the trial. Figure 12 reveals the greater cooling effect of the dry ice compared to the ice pack for the first day but the difference between the dry ice (DI) and ice pack (IP)-groups decreases during the storage as seen in Figure 11. The greater cooling effect of the dry ice compared to the ice pack during the first few hours of such cold chains has already been shown by Magnusson et al. (2009).



**Figure 12. Product temperature for the first day after packaging at the processing plant during land transport to Matis, where it was stored in an air climate chamber. Left: boxes sampled on day 10.**

**Right: boxes sampled on day 13. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.**

The mean product temperature in the two LC-DI boxes during the first day of the trial was calculated as  $(-0.9 \pm 0.2)$  °C for the both boxes (sampled on d10 and d13). The corresponding mean product temperature in the SIC-DI boxes during the first day of the trial was  $(-1.2 \pm 0.2)$  °C and  $(-0.9 \pm 0.5)$  °C for the boxes sampled on d10 and d13, respectively. This indicates that the lower cooling medium temperature of the slurry ice (SIC) depicted in Figure 9 than of the liquid (LC) shown in Figure 8, results in slightly lower product temperature in the beginning of the cold chain. Product temperature after packaging between  $-1.0$  and  $-0.5$  °C should contribute to increased quality and prolonged shelf life of the cod fillets compared to product temperature around  $0$  °C. Thus, the slurry ice is preferred as a pre-cooling medium for fish fillets, at least in case of problems regarding temperature control of the liquid.

### **3.2 Sensory evaluation**

Figure 13 shows how the sensory characteristics of the sample groups change with storage time. The first and second principal components explain 88 and 5% of the variation between samples. At the beginning of storage, all samples are mainly described by sweet odour and flavour, metallic flavour, shellfish odour and juicy texture. As the storage time progresses, these characteristics become less evident, the samples become more described by meaty odour and flavour and then odour of boiled potatoes. At the end of store, the samples become increasingly described by spoilage attributes, such as table cloth odour, pungent flavour, off flavour, TMA and sour odours and flavours (Figure 13b). These changes appeared to occur faster in the LC-IP group compared to the other three groups (Figure 13a).

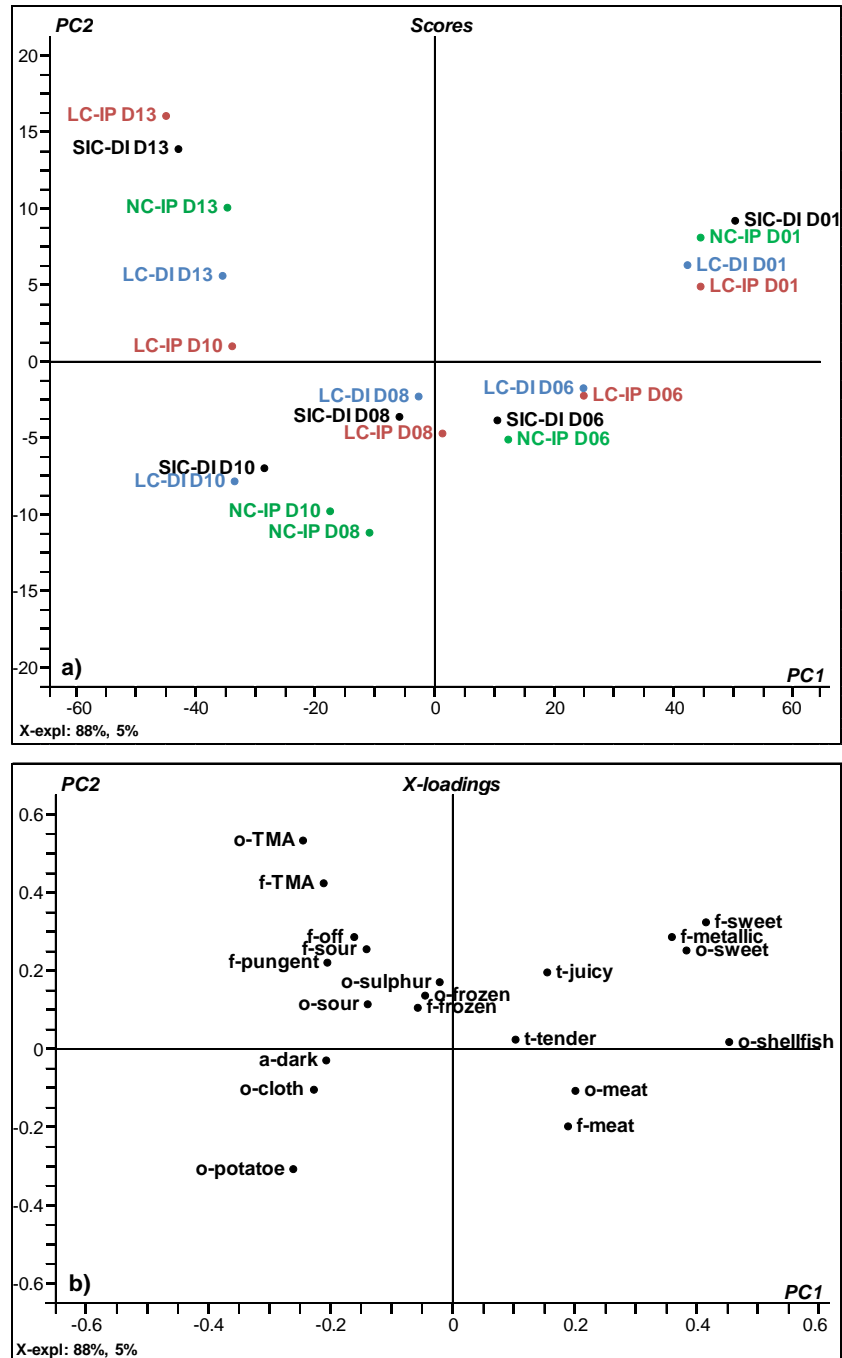


Figure 13. PCA describing sensory quality, odour (o-), appearance (a-), flavour (f-) and texture (t-) of the sample groups with storage time (d). PC1 VS PC2 (X-expl.: 88% and 5%). a) scores, b) X-loadings.

Figure 14 shows the Torry freshness scores of the groups with storage time. A Torry score around 7 indicates the fish has lost most of its freshness odour and flavour characteristics, and has a rather neutral odour and flavour (Shewan et al 1953). After six

days of storage, all groups were above or around these limits, but on storage day eight, all groups were below these limits. When the average Torry score is around 5,5 most of the sensory panellists detect spoilage attributes, and these limits have been used as the limits for consumption at Matis (see e.g. Olafsdottir and others 2006). According to this, the maximum shelf life of LC-IP was 9 days, 10-11 days for SIC-DI and LC-DI and 11-12 for NC-IP.

Figure 15 shows how the sweet flavour changed with storage time. When the score for this attribute is around 25-30, the fish has lost most of its characteristic sweet flavour. After eight days, all groups were around these limits. This is slightly longer than what was observed with the Torry method.

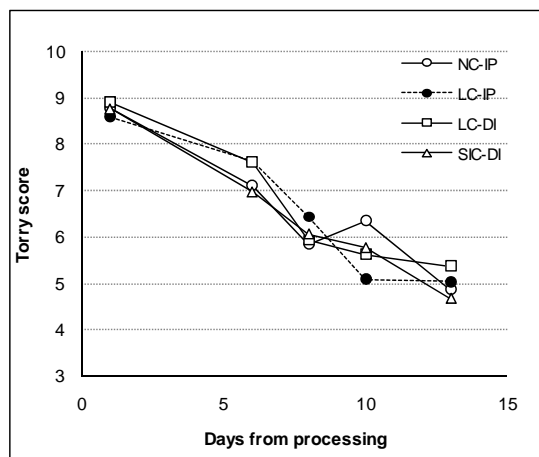


Figure 14. Average Torry freshness scores.

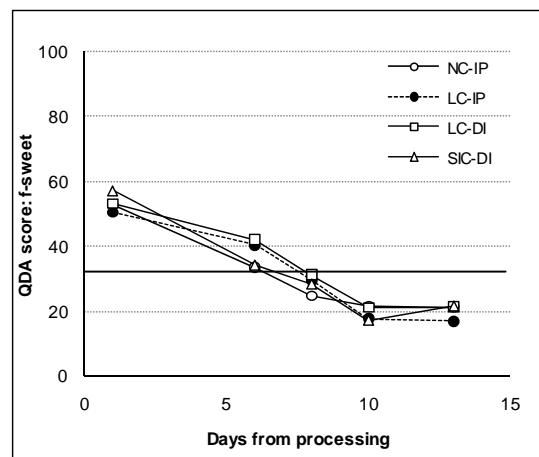


Figure 15. Average QDA scores of sweet flavor.

Figure 15 - Figure 19 show how odour and flavour attributes related to spoilage change with storage time. A part of the panel could not taste the samples due to spoilage, which explains the inconsistency between the intensity of TMA odour and flavour. End of shelf life is usually determined when sensory attributes related to spoilage become evident. When the average QDA score for those attributes is above the value 20 (on the scale 0 to 100) most panellists detect them (Bonilla and others 2005; Magnússon and others 2006). According to this criterion, the maximum shelf life of LC-IP and LC-DI was 10 days, SIC-DI and NP-IP 13 days.

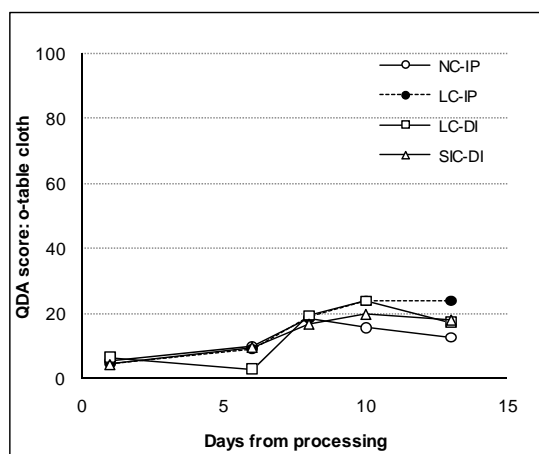


Figure 16. Average QDA scores of table cloth odour.

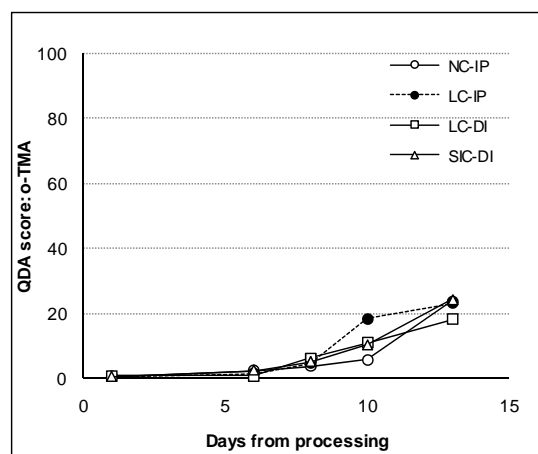


Figure 17. Average QDA scores of TMA odour.

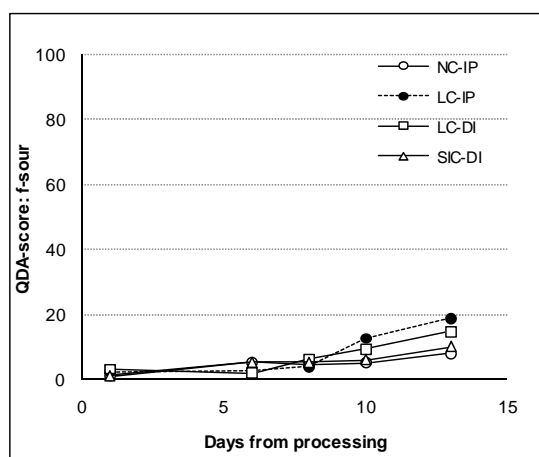


Figure 18. Average QDA scores of sour flavor.

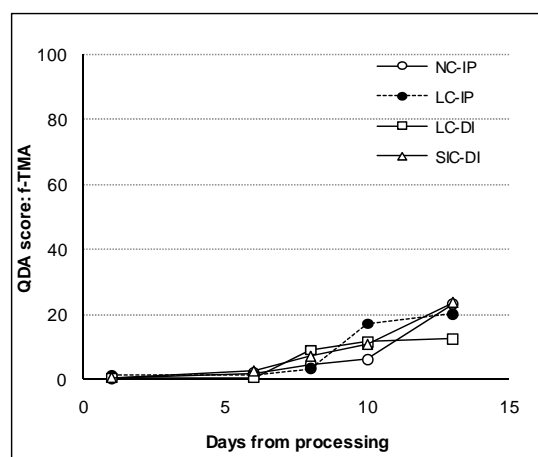


Figure 19. Average QDA scores of TMA flavor.

Table 5. Freshness period and maximum shelf life of cod fillets (days from processing) according to sensory evaluation.

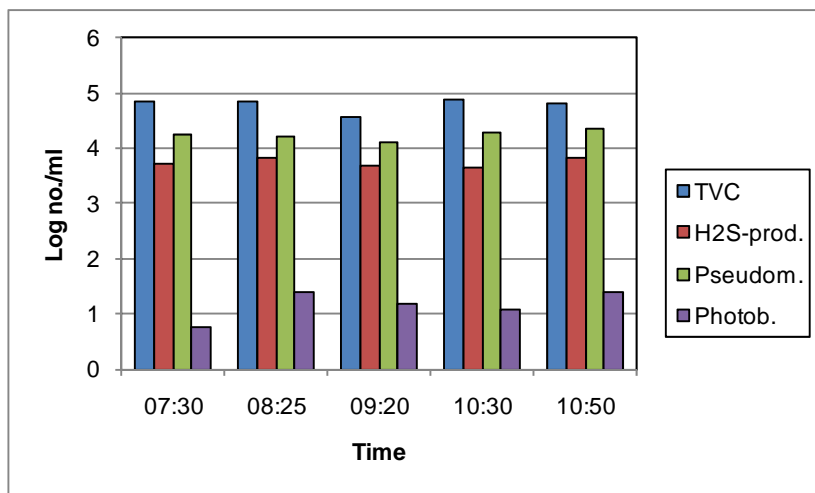
Group*	Freshness period	Shelf life
NC-IP	6-8	11-13
LC-IP	6-8	9-10
LC-DI	6-8	10-11
SIC-DI	6-8	11-13

\*The cod was caught 3 days before processing.

The different treatments of the groups did not influence the sensory characteristics of the samples in other ways than resulting in different maximum shelf life. The estimation of these periods was based on freshness and spoilage related odour and flavour attributes.

### 3.3 Microbial measurements

Initial microbial counts in the liquid cooling brine on the day of processing (3 day post-catch) are shown in Figure 19. Considerable load of microbes was found in the brine, including H<sub>2</sub>S-producing bacteria. It is very probable that the higher microbial counts found in the fillets that were immersed into the cooling liquid brine compared to the control fillets (no cooling) and fillets that were cooled in slurry ice may be attributed to this. It is clear that the fillets have picked up microbes from the liquid brine. However, hardly any increase in microbes was observed in the brine during the time period.



**Figure 20. Total viable counts (TVC), H<sub>2</sub>S-producing bacteria, presumptive pseudomonads and *Photobacterium phosphoreum* in the liquid brine on the day of processing.**

Results from microbial counts are shown in Figures 20-21. In all cases, highest counts were seen in the group where liquid cooling was applied after skinning and ice packs used during packaging (LC-IP). The other group where liquid cooling was applied and dry ice used during packaging (LC-DI) was most often the second highest in microbial counts. Counts in the groups with no cooling-ice packs (NC-IP) and slurry ice cooling-dry ice (SIC-DI) were generally lowest.



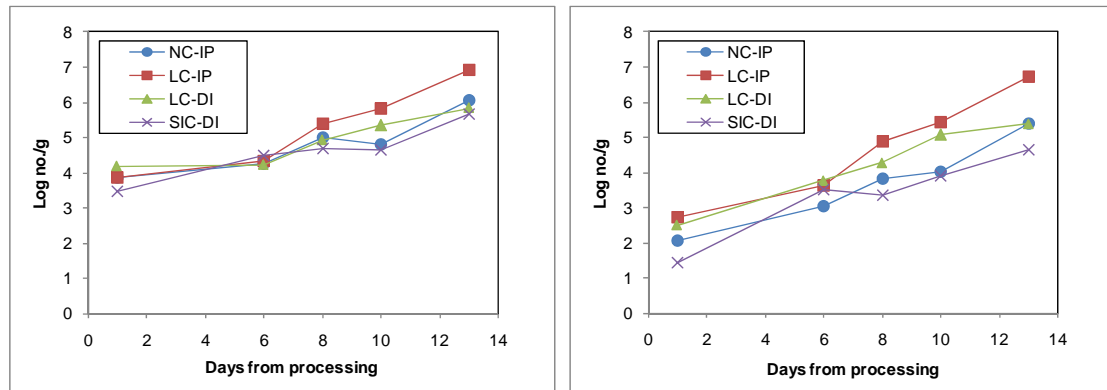


Figure 21. Total viable counts (left) and counts of  $H_2S$ -producing bacteria (right) in cod fillets. Mean of two samples. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.

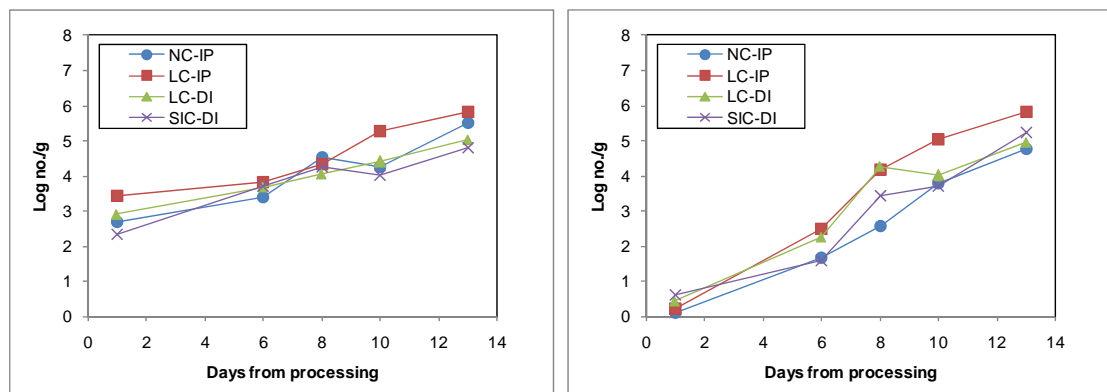


Figure 22. Growth of presumptive pseudomonads (left) and *Photobacterium phosphoreum* (right) in cod fillets. Mean of two samples. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.

### 3.4 Chemical measurements

#### 3.4.1 Total Volatile Base Nitrogen and Trimethylamine

Results from TVB-N and TMA measurements are shown in Figure 22. Highest values at the end of storage (day 13 from processing) were seen in the group where liquid cooling was applied after skinning and ice packs used during packaging (LC-IP). The other group where liquid cooling was applied and dry ice used during packaging (LC-DI) was second highest. TVB-N and TMA values were lowest in the groups where no cooling-ice packs (NC-IP) and slurry ice cooling-dry ice (SIC-DI) was applied. These results are in good harmony with the results obtained from microbial counts.

The rapid increase in both TVB-N and TMA in the later stages of the storage period in groups LC-IP and LC-DI may be largely attributed to rapid growth of especially  $H_2S$ -

producing bacteria (presumably *Shewanella putrefaciens*) but also *Photobacterium phosphoreum* in these groups but both these bacteria are active reducers of trimethylamine oxide (TMAO) to TMA.

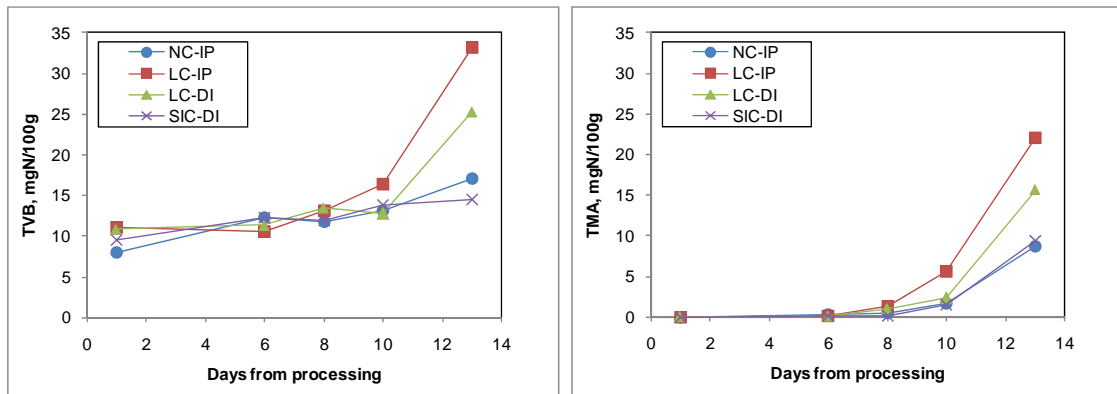


Figure 23. Total Volatile Base Nitrogen (TVB-N-left) and trimethylamine (TMA-right) in cod fillets. Mean of two samples. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.

### 3.4.2 pH - measurements

Results from pH measurements are shown in Figure 23. In all experimental groups, pH was in the narrow range of 6.7 - 7.0 over the storage period. Highest pH values might have been expected in the group LC-IP at the end of storage where TVB-N and TMA were highest. That was however not the case.

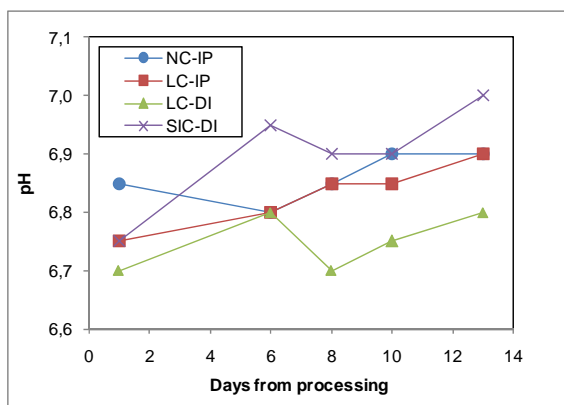


Figure 24. pH in cod fillets. Mean of two samples. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.

### 3.4.3 Salt and water content

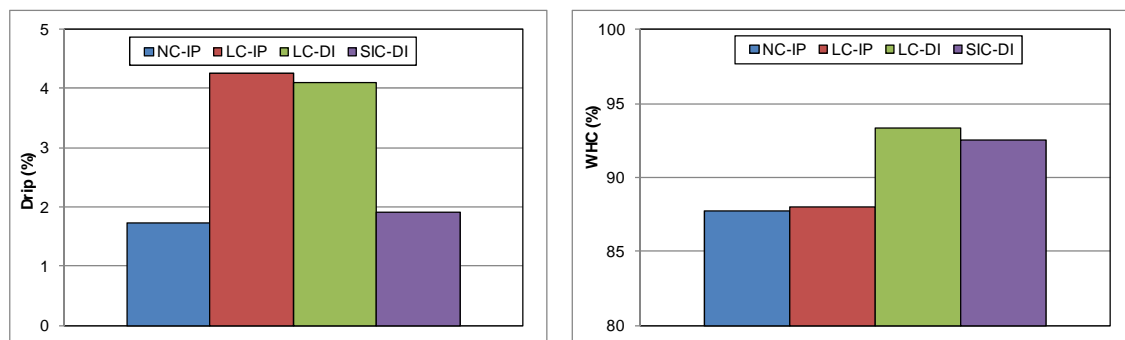
The salinity of the slurry ice was 0.7% (sample taken at 09:25 AM). The salinity of the liquid in the liquid cooling tank was between 1.5% (sample taken at 10:50 AM) and 2.2% (sample taken at 07:30 AM).

Salt content was 0.2 - 0.3% in non-precooled fillets (NC-IP) but 0.4% in fillets precooled in liquid brine (LC-IP and LC-DI) and 0.3% in fillets precooled in slurry ice (SIC-DI).

The water content for the four groups was between 80.7% and 81.1% as measured on day 14 after processing.

### 3.5 Water holding capacity and drip

Figure 25 shows the drip and water holding capacity for the four experimental groups as measured fourteen days after processing. Greater drip was observed for the two liquid cooling (LC)-groups compared to the no cooling (NC) and slurry ice cooling (NC) groups. This is in contrast to the results of Magnusson et al. (2009), who found very similar drip for non-pre-chilled fillets on one hand and liquid-pre-chilled fillets on the other hand. However, it should be noted, that only one measurement on d14 was conducted in the current study. The same applies for the water holding capacity; it was only measured on d14 (Figure 25, right). Higher water holding capacity was observed for the DI-groups (both for LC and SIC) than for the IP-groups.



**Figure 25. Drip (left) and water holding capacity (right) as measured on day 14 from processing. NC: No cooling, LC: Liquid cooling, IP: Ice packs, SIC: Slurry ice cooling, DI: Dry ice.**

## **4 CONCLUSION**

Results from sensory, microbial and chemical analysis all showed that immersing the skinless cod fillets in liquid cooling brine prior to packaging resulted in reduced shelf life in comparison with fillets that were not immersed in liquid brine (no cooling) or in slurry ice. This is attributed to the fact that the cooling liquid brine carried considerable loads of microbes. Highest microbial and chemical values and shortest shelf life were seen in the experimental groups where liquid cooling was applied after skinning. The results indicate that it is better to use dry ice at packaging than ice packs for liquid cooled cod fillets. If the intention is to cool fillets in liquid brine prior to packaging it must be ensured that the brine is of high microbial quality in order to avoid the danger of cross-contamination from brine to fillet. Results from temperature measurements indicated that slurry ice is recommended as a pre-cooling medium during processing because of its higher cooling capacity than of the liquid brine.

## **5 ACKNOWLEDGEMENTS**

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