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# Effect of superchilled processing of whole whitefish – pre-rigor

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# **Report summary**



Titill / Title	Effect of superchilled processing of whole whitefish – pre-rigor / Áhrif ofurkælingar á skemmdarferla og geymsluþol heils fisks og flaka										
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Ágrip á íslensku:	geymslu fljótlega ofurkæl hitamæ tilrauna	uþol heils fisks og fl a eftir veiði og e dum þorski og þor lingar, skynmat, e hópa saman, en þei	aka. Rannsókn var gerð á einnig á áhrifum ofurka rski kældum í ís á hefðb efna- og örverumælinga ir voru geymdir við –1.4 til	elingar á skemmdarferla og heilum þorski ofurkældum elingar á flök unnum úr undinn hátt. Notaðar voru r til að bera eftirfarandi –1.2°C meðalhita:							
	1)	NC: heill þorskur k									
	2)	SC: ofurkældur hei	•								
	3)		n flakavinnsla úr heilum þ								
	4)		ök unnin úr heilum þorski								
	5)		n flakavinnsla úr ofurkældi								
	6)	SC-SC: ofurkæld flo	ök unnin úr ofurkældum h	eilum þorski							
	Niðurstöður skynmats benda til þess að ofurkæld vinnsla á heilum þorski geti lengt geymsluþol hans um tvo daga. Ofurkæling á heilum þorski hafði ekki áhrif á sýrustig, vatnsinnihald, vatnsheldni og örveruvöxt í heilum fisk samanborið við fisk sem ekki var ofurkældur í vinnslu. Samkvæmt skynmati var lítinn mun að finna á geymsluþoli mismunandi flakahópa. Geymsluþol var metið 16–18 dagar, sem er nokkuð langur tími fyrir þorskflök. Ferskleikatímabil tilraunahópsins SC-SC virtist þó vera heldur lengra en hinna hópanna. Líkt og fyrir heila þorskinn reyndist lítill munur milli flakahópanna m.t.t. örveruvaxtar, efna- og eðliseiginleika. Takmarkaðan mun milli tilraunahópa má mögulega skýra með stöðugum og ofurkældum geymsluaðstæðum. Með hliðsjón af því er ráðgert að framkvæma aðra sambærilega tilraun þar sem hermt verður eftir dæmigerðari umhverfishitaferlum í flutningi ferskfiskafurða (0–4 °C) en í þessari tilraun (–1.4 til –1.2 °C).										
Lykilorð á íslensku:	heill þo	rskur, ofurkæld vi	nnsla, geymsluþol, sken	nmdarbakteríur							

# Skýrsluágrip Matís ohf

Icelandic Food and Biotech R&D

### **Report summary**

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#### Summary in English:

The main aim of the study was to study the effects of superchilled processing on storage life of both whole fish and fillets. The following experimental groups were evaluated by means of temperature monitoring, chemical- and microbial measurements and sensory evaluation, which were stored at mean temperatures of -1.4 to -1.2 °C:

- 1) NC: non-superchilled whole cod
- 2) SC: superchilled whole cod
- 3) NC-NC: non-superchilled fillets from non-superchilled whole cod
- 4) NC-SC: superchilled fillets from non-superchilled whole cod
- 5) SC-NC: non-superchilled fillets from superchilled whole cod
- 6) SC-SC: superchilled fillets from superchilled whole cod

The results from the sensory evaluation indicate that superchilled processing of whole cod can extend shelf life by two days. Differences in values of pH, water content, water holding capacity and bacterial growth between the superchilled and non-superchilled whole fish groups were minor. Differences in sensory scores between the fillet groups were small. Shelf life was estimated between 16 and 18 days which is quite long shelf life for cod fillets. However, the group SC-SC seemed to retain freshness a little longer than other groups. As in case of the whole cod, the differences in bacterial count, chemical and physical properties between the fillet groups were small. Very similar fish temperatures between both the whole fish and the fillets groups resulting from the superchilled storage conditions applied may be the main reason for the small differences obtained. Thus, another study with more common temperature conditions during transport and storage of fresh fish (chilled but not superchilled) will be performed.

English keywords:

whole fish, superchilled processing, shelf life, spoilage bacteria

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#### 1 Introduction

Superchilling of fresh food implies that the food temperature is lowered to no more than 1–3 °C below the initial freezing point of the food (Aune, 2003; Magnussen, 2008; Kaale et al., 2011). The initial freezing point of cod is specified by Rahman (2009) as –0.9 °C. The term superchilling has also been used when chilling foods to a temperature only below the freezing point of water, i.e. 0 °C (Aune, 2003; Ando et al., 2004) which generally is higher than the initial freezing point of foods. Superchilling cod with water content of 80–82% down to –2 °C implies that around 50–55% of the fish water content is frozen (Rha, 1975). In order to avoid excessive surface freezing and ice crystal growth causing possible structural damage and negative texture changes to the fish flesh and increase drip loss (Magnussen, 2008; Kaale et al., 2011), cod is normally superchilled to no lower temperature than –1 °C in modern industrial applications (Valtýsdóttir et al., 2010; Stevik and Claussen, 2011).

The SuperChiller cooling technique by Marel Ltd., Garðabær, Iceland (formerly referred to as combined blast and contact (CBC) cooling technique by Skaginn Ltd., Akranes, Iceland) has proven to be an efficient superchilled processing method for fresh whitefish loins/fillets before packaging and prolonging storage life due to inhibited microbial activity (Martinsdóttir et al., 2005; Gao, 2007; Magnússon et al., 2009). Before entering the SuperChiller, fish fillets are conveyed through a liquid cooler/slurry ice cooler with salinity of approximately 1.0-2.5% which prevents excessive freezing of the fish flesh. Measurements have shown that the required holding time in the SuperChiller is around 8-10 min in order to reduce the fillet temperature from around 1-4 °C to temperatures around -1 °C with the air temperature in the SuperChiller set at -8 °C (Gao, 2007; Valtýsdóttir et al., 2010). Cod temperature of -1 °C corresponds to a frozen water ratio around 10-15% (Rha, 1975). This means that extra energy is needed for melting the partly frozen water, which provides protection against thermal abuse during distribution.

The research project "Ofurkældur heill fiskur - fyrir dauðastirðnun" (e. Super-Chilled Round Fish – Pre Rigor) is funded by AVS R&D Fund of Ministry of Fisheries in Iceland (project no. R 062-11). The main aim of the project is to investigate if superchilled processing and storage of whole whitefish can prolong the storage life of whitefish products even further than by the 2–4 days that superchilled processing and storage of fillets can do (Martinsdóttir et al., 2005; Olafsdottir et al., 2006; Gao, 2007; Magnússon et al., 2009).

#### 2 Material and methods

#### 2.1 Experimental design

#### 2.1.1 Whole fish groups (NC, SC)

The raw material used in the experiments was caught between 4 AM and 8:30 AM on November 24, 2012 by the long liner Kristján HF-100 in Hvalfjörður, Southwest-Iceland. After bleeding in cold seawater the cod was chilled in thin slurry ice prepared from sea and ice (around 50–60 kg of ice with 400–450 kg of fish) in insulated tubs with storage capacity of 660 L. The cod was landed at 10:30 AM in Hafnarfjörður in Southwest-Iceland at ambient temperature of around 2 °C and transported in insulated tubs in a non-insulated truck to the processing plant Eskja less than 1 km from the harbour.

After gutting between 11 and 11:30 AM in the processing plant, the non-superchilled (NC) group was prepared by packing 70 whole fish weighing 127.5 kg in 5 layers with 77 kg of ice in between the fish layers in a 460-L tub. The superchilled (SC) group was prepared by precooling 83 whole fish weighing in total 206 kg in slurry ice at temperature of around -2 °C and salinity of 3% for around 11 min followed by combined blast and contact cooling (Figure 1) at -12 °C for 13–15 min in a SuperChiller (Marel, Garðabær, Iceland). After the superchilling, the fish was packed in 5 layers with 110 kg of ice in between the fish layers in a 460-L tub (Figure 2). At 1:30 PM the two tubs were transported to Matís, where they were stored at around -1 °C in a controllable air climate chamber until day 6 post-catch, i.e. Nov 30. On that day, most of the whole fish was transported back to the processing plant in the insulated tubs for processing while 15 whole fish specimen were kept for further iced and superchilled storage at Matís. Sampling was done on days 1, 4, 6, 11, 14 and 18 post-catch, see Table 1.

Ibutton temperature loggers (DS1922L) from Maxim Integrated Products (Sunnyvale, CA, USA) were used to monitor the temperature both ambient and product temperatures. The ambient temperature was measured at 2–3 positions for each group and the product temperature was measured in 2 and 5 fish in the NC and the SC groups, respectively. The Ibutton temperature loggers had a resolution of 0.0625 °C, measurement range of -40 to 85 °C and an accuracy of  $\pm 0.5$  °C between -15 and 65 °C. All temperature loggers were factory calibrated and re-calibrated by the authors in a thick mixture of fresh crushed ice and water.



Figure 1. Whole cod fish superchilled by means of combined blast and contact cooling.



Figure 2. Superchilled cod packed in ice.

#### 2.1.2 Fillet groups (NC-NC, NC-SC, SC-NC, SC-SC)

The processing was performed on Nov 30, 2012 in the same processing plant as the pre-processing on Nov 24. The experimental groups are presented in Table 1. The superchilled fillets groups (NC-SC and SC-SC) were prepared by slurry ice cooling for 10 min and combined blast and contact cooling in the SuperChiller at -7 °C for 10 min.

After packaging into 3-kg expanded polystyrene (EPS) boxes (Promens Tempra, Hafnarfjörður, Iceland), product and ambient temperatures were monitored for two boxes in each of the four fillet

experimental groups. Two temperature loggers were put in each of the monitored EPS boxes and one on the outside of each box, see Figure 3. Following this, the four fillet experimental groups were transported back to Matís for further storage at -1 °C in the same air climate chamber as was used for the whole fish.

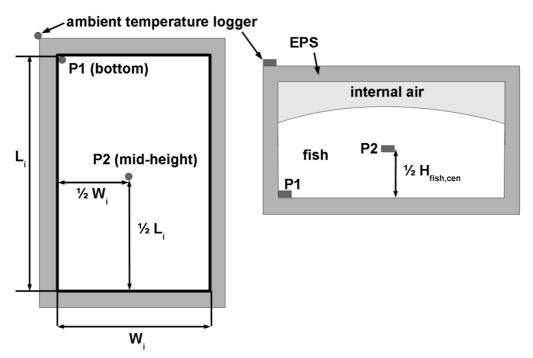


Figure 3. Positions of ambient and product temperature loggers on the outside and inside the temperature monitored EPS boxes in a horizontal plane (left) and in a vertical plane (right).

Sampling was done on days 7, 11, 14 and 18 post-catch, which corresponds to days 1, 5, 8 and 12 post-processing, see Table 1.

Table 1. Overview of the experimental groups and the corresponding sampling days.

Group	Description	Sampling days (post-catch)
NC	Non-superchilled whole fish	1, 4, 6, 11, 14, 18
SC	Superchilled whole fish	1, 4, 6, 11, 14, 18
NC-NC	Non-superchilled whole fish, non-superchilled fillets	7, 11, 14, 18
NC-SC	Non-superchilled whole fish, superchilled fillets	7, 11, 14, 18
SC-NC	Superchilled whole fish, non-superchilled fillets	7, 11, 14, 18
SC-SC	Superchilled whole fish, superchilled fillets	7, 11, 14, 18

#### 2.2 Sensory evaluation

Sensory evaluation of the whole cod was carried using the Quality Index Method (QIM) (Martinsdóttir et al 2001). Cooked samples were evaluated using profiling method (generic descriptive analysis, DA) where the intensity of sensory attributes was evaluated to describe sensory characteristics in flavour, odour, appearance and texture (Stone and Sidel, 1985). Cooked samples

were also evaluated with a Torry freshness scale (based on a method to evaluate the freshness of lean fish described by Shewan et al. (1953). Whole cod was evaluated with QIM, Profiling method and Torry freshness scale on day 1, 4, 6, 11, 14 and 18 and fillets on day 7, 11, 14 and 18. Eleven trained panellists all experienced in sensory evaluation participated in the sensory evaluation. For evaluation with QIM, three fish of each sample group were coded with three digit numbers and laid on a table. The fish were then evaluated individually by the panellists. For profiling method a scale from previous experiments on cod was used (Margeirsson et al., 2011). The scale includes 26 sensory attributes which are listed in Table 2. The intensity of each attribute for a given sample was described using an unstructured scale (from 0–100). The samples were 40–50 g of the loin part from a cod fillet. They were steamed for 6 minutes and presented warm to the panellists in small aluminium boxes closed with lids. All samples were coded with three digit numbers and presented in a random manner. Three or four samples were evaluated at a time. Each sample was evaluated using the Torry freshness scale immediately after evaluation with profiling method. The sensory evaluation program FIZZ (2.10c, 1994-2005, Biosystémes) was used to collect sensory data.

Table 2. Sensory attributes for boiled cod and their definitions.

sensory attribute	short name	scale	definition
ODOUR			
sweet	O-sweet	none    much	Sweet odour
shellfish, algae	O-shellfish	none    much	Shellfish, algae, characteristic fresh odour
vanilla	O-vanilla	none    much	Vanilla, sweet hot milk
boiled potatoes	O-potatoes	none    much	Reminds of whole warm boiled potatoes
dishcloth	O-dishcloth	none    much	Reminds of a dishcloth (damp cloth to clean kitchen table, left for 36 h)
TMA lykt	O-TMA	none    much	TMA odour, reminds of dried salted fish, amine
spoilage sour	O-sour	none    much	Sour odour, sour milk, spoilage sour, acetic acid
sulphur	O-sulfur	none    much	Sulphur, matchstick
APPEARANCE			
colour	A-colour	light    dark	Left end: light; white colour. Right end: dark; yellowish, brownish, grey
heterogeneous	A-heterog.	homogenous    heterogenous	Left end: homogeneous, even colour. Right end: heterogeneous colour,
white precipitation	A-precipit.	none    much	White precipitation on the sample surface
flakiness	A-flakes	none    much	The fish portion slides into flakes when pressed with the fork
FLAVOUR			
salt	F-salt	none    much	Salty taste
metallic	F-metallic	none    much	Characteristic metallic flavour of fresh cod
sweet	F-sweet	none    much	Characteristic sweet flavour of fresh (boiled) cod
pungent	F-pungent	none    much	Pungent flavour, bitter
spoilage sour	F-sour	none    much	Sour taste, spoilage sour
TMA	F-TMA	none    much	TMA flavour, reminds of dried salted fish, amine
putrid	F-putrid	none    much	Putrid
TEXTURE			
soft	T-soft	firm    soft	Softness in first bite
juicy	T-juicy	dry    juicy	Dry - draws liquid from the mouth
tender	T-tender	tough    tender	Evaluated after chewing several times
mushy	T-mushy	none    much	Mushy texture, puree
meaty mouthfeel	T-meaty	none    much	Meaty texture, meaty mouthfeel, crude muscle fibers
astringent	T-astringent	none    much	Astringent, dry red wine, tannin
rubbery	T-rubbery	none    much	Rubbery texture, springy

#### 2.3 Microbial measurements

Minced flesh (20 g) was mixed with 180 g of chilled Maximum Recovery Diluent (MRD, Oxoid, UK) in a stomacher for 1 minute. Successive 10 fold dilutions were done as required. Total viable psychrotrophic counts (TVC) were performed on iron agar (IA) as described by Gram et al. (1987) with the exception that 1% NaCl was used instead of 0.5% with no overlay. Counts of H<sub>2</sub>S producing bacteria (black colonies) were evaluated on IA. Plates were spreadplated and incubated at 17 °C for 5 days. Enumeration of presumptive pseudomonads was performed using modified Cephaloridine Fucidin Cetrimide (mCFC) agar as described by Stanbridge and Board (1994). *Pseudomonas* Agar Base (Oxoid, UK) with CFC selective Agar Supplement (Oxoid) was used and the Plates were spreadplated and incubated at 22 °C for 3 days. Mean bacterial numbers are presented as log10 numbers of colonyforming units (cfu) per gram fish.

#### 2.4 Chemical and physical measurements

#### 2.4.1 Total Volatile Base Nitrogen

The method of Malle and Tao (1987) was used for total volatile bases (TVB-N). TVB-N was measured by steam distillation (Struer TVN distillatory, STRUERS, Copenhagen) and titration, after extracting the fish muscle with 7.5% aqueous trichloracetic acid solution. The distilled TVB-N was collected in boric acid solution and then titrated with sulphuric acid solution. All chemical analyses were done in triplicate.

#### 2.4.2 pH-measurements

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

#### 2.4.3 Water content and water holding capacity

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).

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 x g for 5 minutes at 0–5 °C. WHC (%) was then calculated with the following equations:

$$WHC (\%) = \frac{Water \ content \ (\%) - \ Weight \ loss \ (\%)}{Water \ content \ (\%)} \ x \ 100$$

where the weight loss is defined as:

Weight loss (%) = 
$$\frac{Weight loss in centrifuge (g)}{Original sample weight (g)} \times 100$$

#### 2.5 Data analysis

Analysis of variance (ANOVA) was carried out on QIM, Torry and profiling 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

#### 3A. Whole fish

#### **3A.1 Temperature measurements**

The mean ambient and product temperatures for the two whole fish groups are presented in Table 3. Clearly, very similar temperatures were measured both for the two groups and their ambience. The ambient temperatures measured at four different positions outside the fish tubs/boxes containing the NC/SC-fish are shown in Figure 4. The short peaks seen in the graph are caused by short openings of the cooling chambers during sampling. The ambient temperature is presented on a more narrow scale in Figure 5.

Table 3. Mean ambient and product temperatures with  $\pm$  one standard deviation during storage of whole cod for 18 days.

Group	Ambient temperature (°C)	Product temperature (°C) (18 days)	Product temperature (°C) (first 6 days)
NC	-1.4 ± 0.9	-1.2 ± 0.2	-0.8 ± 0.1
SC	-1.3 ± 0.8	-1.2 ± 0.2	-1.0 ± 0.1

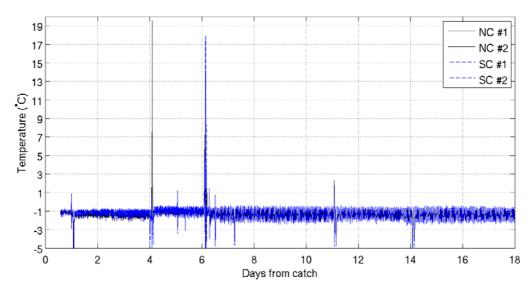


Figure 4. Ambient temperature during storage of whole cod; superchilled (SC) and non-superchilled (NC).

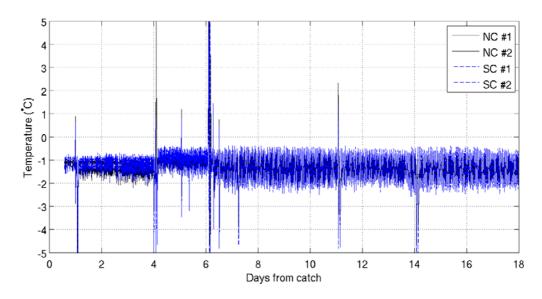


Figure 5. Zoom-up of ambient temperature during storage of whole cod; superchilled (SC) and non-superchilled (NC).

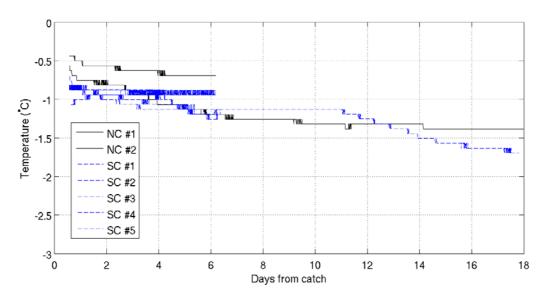


Figure 6. Product temperature during storage of whole cod; superchilled (SC) and non-superchilled (NC).

The product temperatures measured in two NC-fish and five SC-fish is shown in Figure 6. Due to technical failure of the temperature loggers, only temperatures after 9 PM on the day of catching, i.e. around 12 hours post-catch, are presented. When the temperature logging started, the fish had already been stored packed in ice at around -1 °C since around 2 PM, i.e. for 7 h. This explains why the highest temperature of the NC-group monitored is around -0.5 °C, but not around 1-2 °C as was measured with handheld thermometers right after gutting in the processing plant. Product temperature was measured in only one NC-fish and one SC-fish throughout all the 18-day storage and the mean product temperatures  $-1.2 \pm 0.2$  presented in Table 3 are calculated from these fish. The slightly higher mean product temperatures during the first 6 days show that the fish temperature in both groups was decreasing throughout the whole storage period.

It should be noted that the missing temperature data from the first 8–9 hours after the preprocessing causes a slight under-estimation of the product temperature difference between the NC and the SC groups during the first days. This means that the relatively fast product temperature decrease during the superchilled processing is not shown in the results. Despite the lack of temperature monitoring during the first 8–9 hours, it can be stated that the superchilled storage conditions resulted in very similar mean product temperatures in the two groups NC and SC.

Figure 7 shows the cooling curves for whole cod during superchilling for 8, 14 and 18 minutes followed by a chilled storage in insulated EPS boxes (Bjarnason, 2012). The experimental setup in the study by Bjarnason (2012) was similar to the current study. Thus it can be assumed that the superchilled whole fish (SC) in the current study was at temperature of around -1 to -0.5 °C 1-2 hours after the superchilled processing.

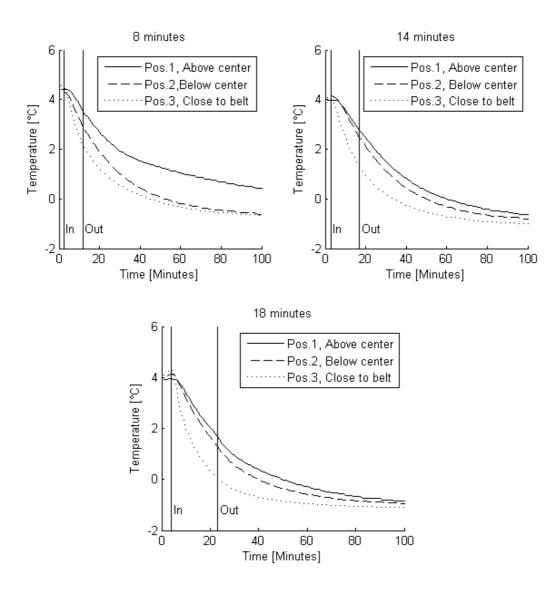


Figure 7. Product temperature of whole cod (weight: 2.5–2.8 kg) fish during superchilled processing at –14.1 °C followed by chilled storage in EPS boxes. The vertical line indicates when the fish were taken out of the superchiller (Bjarnason, 2012).

#### 3A.2 Sensory evaluation

No difference in odour, appearance or flavour was observed between non-superchilled (NC) and superchilled (SC) whole cod before day 6 (Figure 10-Figure 13; table 1 in appendix). On day 11, SC cod was flakier than NC cod. SC cod was also slightly saltier and had more metallic and sweet flavour than NC cod. On day 18, SC cod had more metallic flavour than NC cod. The NC cod had more sour odour than SC cod. No difference was seen in texture on day 1 but difference was marginally significant on day four in mushy and meaty texture. The NC cod had more mushy and less meaty texture. On day six there was a difference in most texture attributes and SC cod was juicier, more tender, more mushy and had less meaty texture than NC cod. No difference was seen in texture on day 11 but on day 14, NC cod was softer, more tender and mushy but had less meaty and astringent texture. This is the opposite of what was seen on day 6. On day 18 similar differences was seen as on day 14. The non-superchilled cod was softer, juicier, more tender and mushy but had less meaty and rubbery texture. Some of the inconsistency between results for different days can be caused by difference between individual fish since the evaluation for whole cod was based on one and a half fish from each group and therefor differences between individuals might affect the evaluation, probably mostly for texture. Sensory evaluation of fillets was based on 6-8 fillets per group which decreases affect from individual difference.

Torry results indicated that the SC cod had two days longer shelf life than the NC cod (Figure 8). Differences between groups were minor during storage time until day 18 where the SC cod scored higher for Torry than the NC cod (p < 0.05). This is supported with profiling results since SC cod in general scored higher on day 18 for freshness attributes such as shellfish odour, metallic and sweet flavour but less for spoilage attributes such as dishcloth odour, TMA, sour and sulfur odour, sour flavour, TMA flavour and putrid flavour even though the difference was not significant apart from sour odour and metallic flavour.

For QI results SC cod received a higher score than NC cod on day 6 but the difference was marginally significant (Figure 9). On day 11 NC cod got a higher score than SC (p < 0.5) but the opposite results were seen on day 18 where SC scored higher than NC cod (p < 0.001).

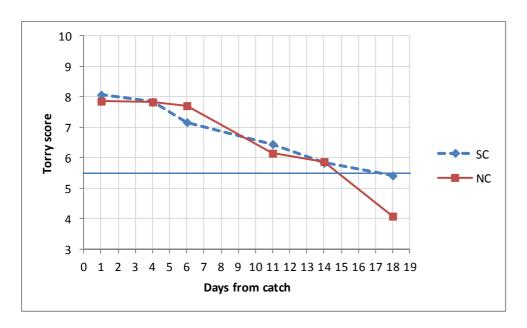


Figure 8. Mean Torry scores for whole cod; superchilled (SC) and non-superchilled (NC).

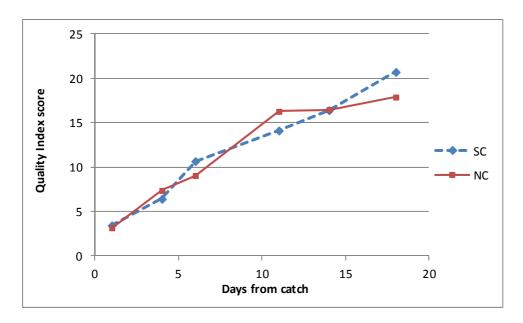


Figure 9. Mean Quality Index (QI) for whole cod; superchilled (SC) and non-superchilled (NC).

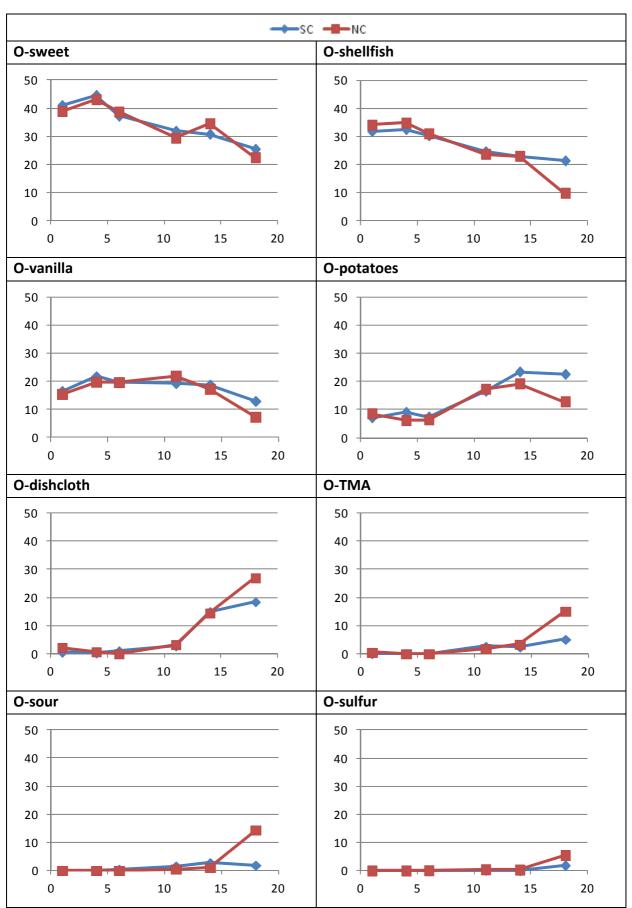


Figure 10. Mean profile scores (0–100, vertical axis) for appearance (O-) attributes for whole cod; superchilled (SC) and non-superchilled (NC). Horizontal axis – days from catch.

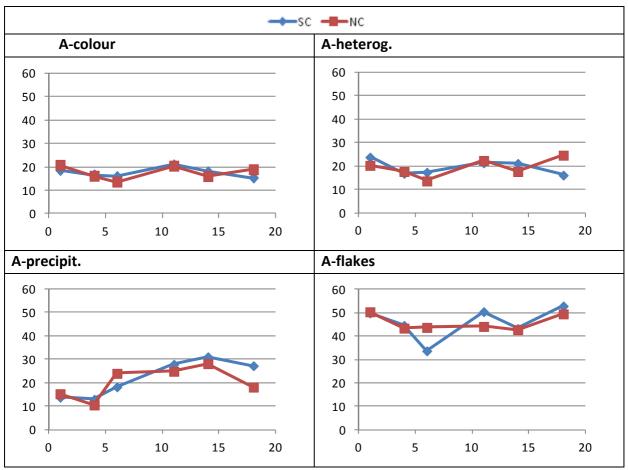


Figure 11. Mean profile scores (scale 0–100, vertical axis) for appearance (A-) attributes for whole cod; superchilled (SC) and non-superchilled (NC). Horizontal axis – days from catch.

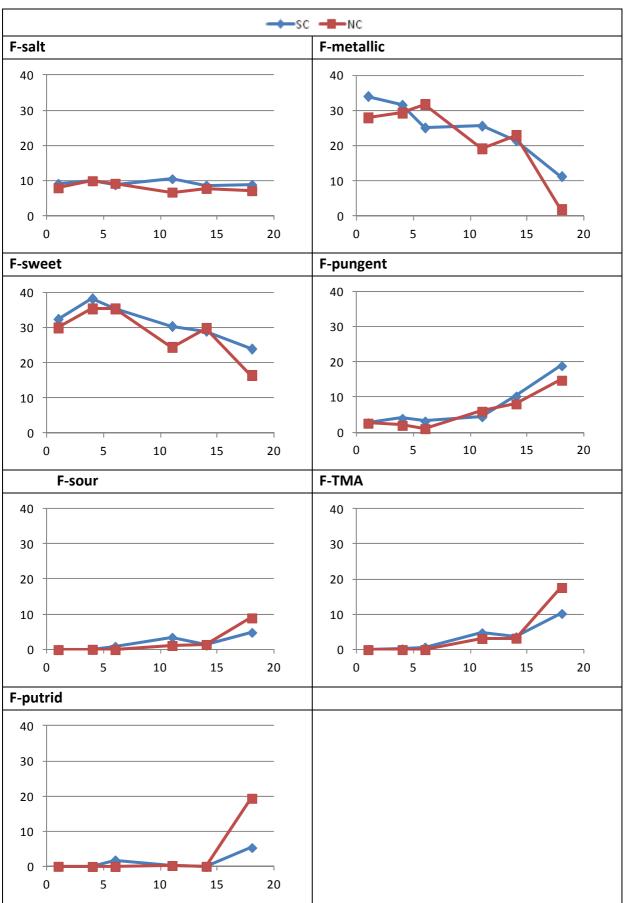


Figure 12. Mean profile scores (scale 0–100, vertical axis) for flavour (F-) attributes for whole cod; superchilled (SC) and non-superchilled (NC). Horizontal axis – days from catch.

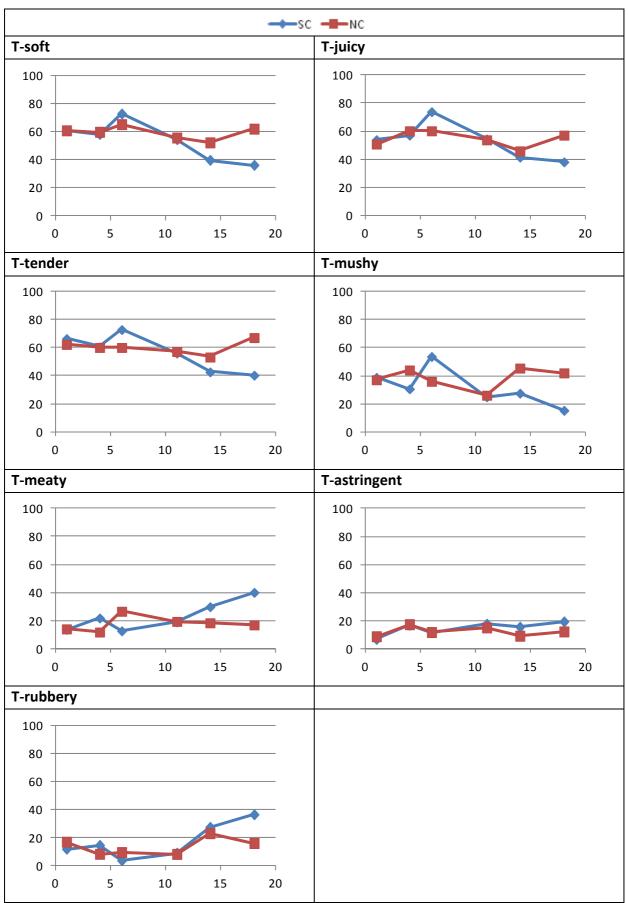


Figure 13. Mean profile scores (scale 0–100, vertical axis) for texture (T-) attributes for whole cod; superchilled (SC) and non-superchilled (NC). Horizontal axis – days from catch.

#### 3A.3 Microbial measurements

Small differences in bacterial growth were seen between non-superchilled and superchilled cod during storage time. On day 1 non-superchilled cod had a higher total viable (TVC) count than superchilled cod but neither H<sub>2</sub>S producing bacteria nor pseudomonads were detected that day. On day 4 the TVC was lower than on day 1 which is sometimes observed during fish storage at low temperatures. No pseudomonads or H<sub>2</sub>S producing bacteria were detected on day 4. On day 6 TVC was higher in superchilled than in non-superchilled cod and the same trend was seen for H<sub>2</sub>S producing bacteria but pseudomonads count was higher in non-superchilled cod. On day 11 no difference in TVC and H<sub>2</sub>S producing bacteria was observed between the groups but a higher count of pseudomonas bacteria was found in superchilled cod than in non-superchilled cod. The pseudomonads count remained about half a log unit higher in superchilled cod throughout the storage time. On day 14 TVC was higher in superchilled cod but no difference was seen between groups for count of H<sub>2</sub>S producing bacteria. On day 18 total viable count and especially count of H<sub>2</sub>S producing bacteria was higher in non-superchilled cod than superchilled cod. Pseudomonads count remained higher in superchilled cod.

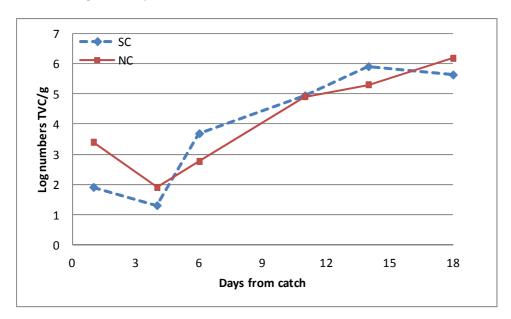


Figure 14. Total viable count per gram on iron agar. Whole cod; superchilled (SC) and non-superchilled (NC).

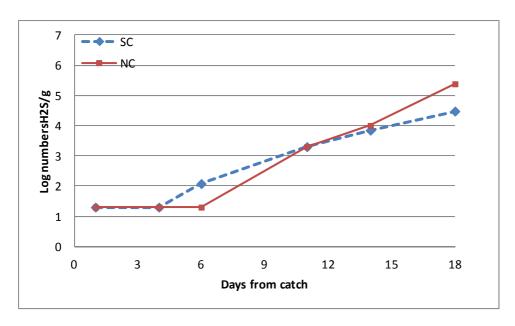


Figure 15. Growth of H<sub>2</sub>S producing bacteria per gram on iron agar. Whole cod; superchilled (SC) and non-superchilled (NC).

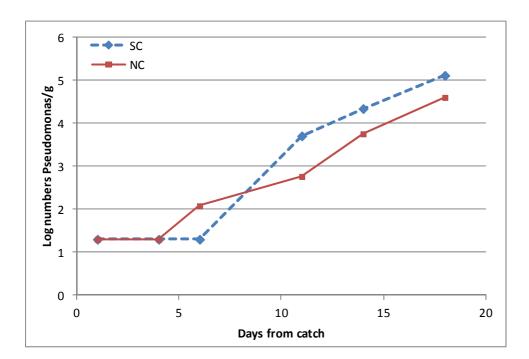


Figure 16. Growth of *Pseudomonas* bacteria per gram on CFC agar. Whole cod; superchilled (SC) and non-superchilled (NC).

#### **3A.4 Chemical and physical measurements**

#### **3A.4.1 Total Volatile Base Nitrogen**

Levels of TVB-N increased slowly during storage but still remained low (Figure 17). No difference was seen between groups.

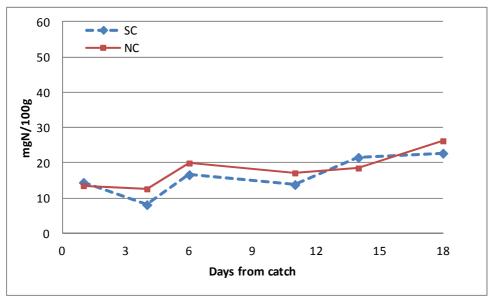


Figure 17. TVB-N values for whole cod; superchilled (SC) and non-superchilled (NC).

#### 3A.4.2 pH-measurements

Non-superchilled cod had in general a higher pH value than superchilled cod except on day 4 where opposite results are seen (Figure 18). Differences in pH values between groups are minor.

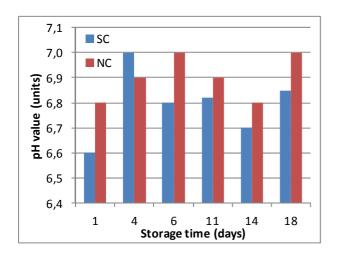


Figure 18. pH values in cod mince (ave ± stdev, n=3). Whole cod; superchilled (SC) and non-superchilled (NC).

#### **3A.4.3 Water content and Water Holding Capacity**

On day 1 the non-superchilled cod had more water content than the superchilled cod, similar results were seen on day 11 but superchilled fish had more water content on day 18. Water holding capacity was generally more in superchilled cod except on day 18 where non-superchilled cod had more water holding capacity than superchilled cod.

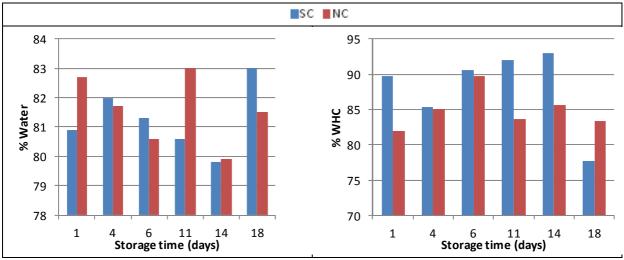


Figure 19. Water content (left) and water holding capacity (WHC-right) in cod mince (ave ± stdev, n=3). Whole cod; superchilled (SC) and non-superchilled (NC).

#### 3B. Fillets

#### **3B.1 Temperature measurements**

The mean ambient and product temperatures for the four fillet groups are presented in Table 4. As in the case of whole fish, very similar product and ambient temperatures were measured for the different groups. The ambient temperatures measured at the outside EPS box surfaces are shown in Figure 20.

Table 4. Mean product temperatures with ± one standard deviations during storage of cod fillets for 12 days.

Group	Ambient temperature (°C)	Product temperature (°C)
NC-NC	-1.4 ± 0.7	-0.8 ± 0.10
NC-SC	-1.2 ± 1.1	-1.0 ± 0.03
SC-NC	-1.4 ± 0.8	-0.8 ± 0.04
SC-SC	-1.3 ± 1.0	-0.8 ± 0.01

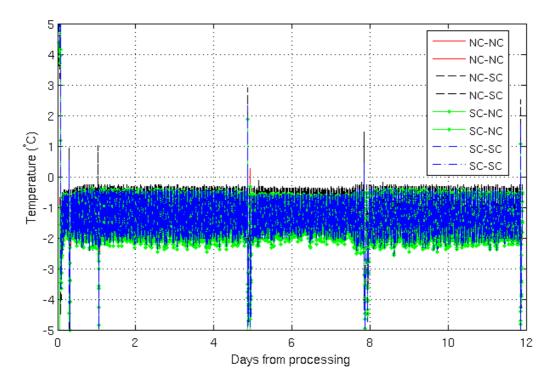


Figure 20. Ambient temperatures during 12-day storage of cod fillets.

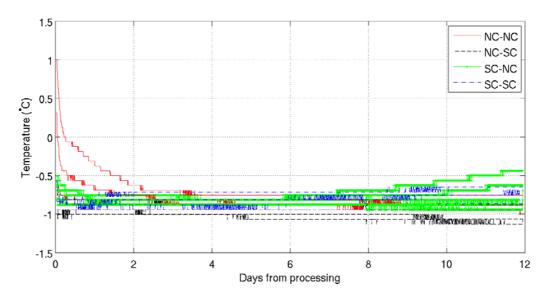


Figure 21. Fillet temperature during 12-day storage post processing.

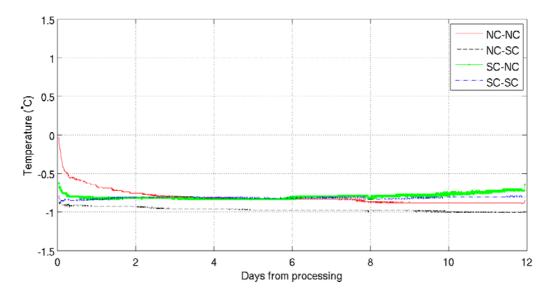


Figure 22. Mean fillet temperatures during 12-day storage post processing.

#### **3B.2 Sensory evaluation**

The cod was filleted on day six. Only two groups were evaluated on day 7; SC-NC and SC-SC but all four groups; NC-NC, NC-SC, SC-NC and SC-SC were evaluated on days 11, 14 and 18. Results are shown in Figure 23—Figure 26 and Table 2 in appendix. On day 7 the difference in sweet odour and white precipitation was marginally significant and SC-SC had sweeter odour and more precipitation but the difference was minimal.

On day 11 no difference was seen between groups in odour and appearance. Difference in metallic flavour was marginally significant and SC-NS had the most metallic flavour but NC-NC the least. Group NC-NC had mushier texture than group SC-SC. Some difference between groups was seen on day 14, especially in odour and appearance. Group SC-SC differed from other groups with sweeter odour, more shellfish and vanilla odours than other groups. Group SC-SC also had less dishcloth odour than groups NC-SC and SC-NC and less TMA odour than group NS-SC. Group NC-SC had more potato odour than groups NC-NC and SC-SC. Group NC-SC was darker than groups NC-NC and SC-SC and had more heterogeneous colour than other groups. Difference in white precipitation and flakes was marginally significant and group NC-SC had the most precipitation but NC-NC the least flakes. Difference in sweet flavour was marginally significant and group SC-SC had the sweetest flavour. Group NC-SC had more TMA flavour than other groups which still was only a trace. Difference in rubbery texture was marginally significant and group SC-NS had the least rubbery texture but group SC-SC the most. On day 18 spoilage attributes were detected in all groups. Some difference was seen between groups. Group NC-NC had the highest score for sweet odour and had a sweeter odour than group SC-SC. Group SC-NC had the highest score for shellfish odour and more than group NC-SC. Group NC-SC had less vanilla odour than other groups. TMA odour was marginally significant and group NC-SC had the most TMA odour but group NC-NC the least. Group SC-SC was lighter in colour than group NC-SC. Group SC-NC had more metallic flavour than group NC-SC. Mushy texture was marginally significant and group SC-SC had the least mushy texture.

Differences in Torry scores between groups were small (Figure 27). Shelf life is estimated between 16 and 18 days which is quite long shelf life for cod fillets. Shelf life of whole fresh cod stored in ice has been reported to be 10–16 days (Jorgensen et al., 1988; Magnússon and Martinsdóttir, 1995; Martinsdóttir et al., 2001). The shelf life of fresh cod fillets stored at 0–1 °C has been reported as 10–12 days (Magnússon and Martinsdóttir, 1995), but up to 17 days if the temperature is kept at –2 °C (Wang et al., 2008).

Groups SC-NC and NC-SC might have a little shorter shelf life than groups SC-SC and NC-NC but group SC-SC seems to retain freshness a little longer than other groups.

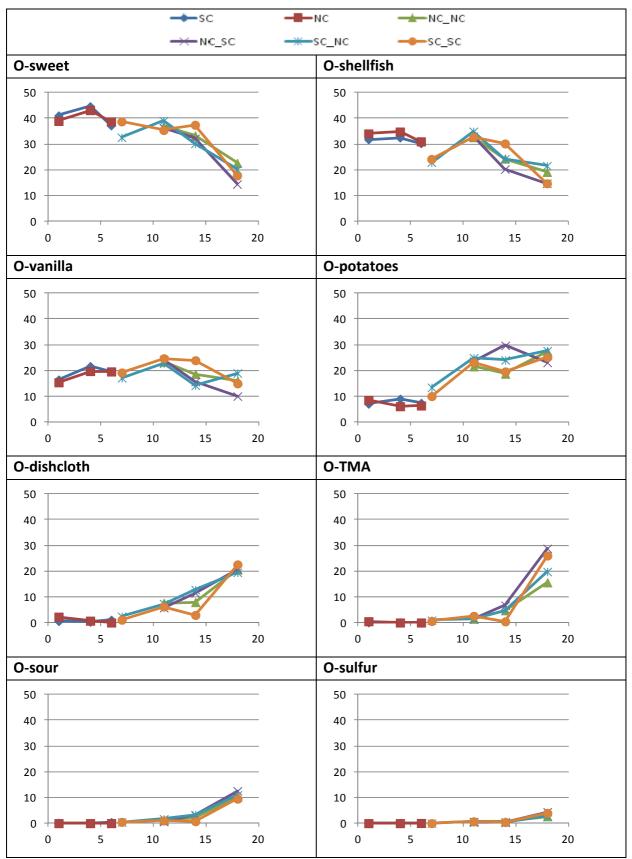


Figure 23. Mean profile scores (scale 0–100, vertical axis) for odour (O-) attributes for whole cod on days 1, 4 and 6: Superchilled (SC) and Non-superchilled (NC) and cod fillets on days 7, 11, 14 and 18: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC). Horizontal axis – days from catch.

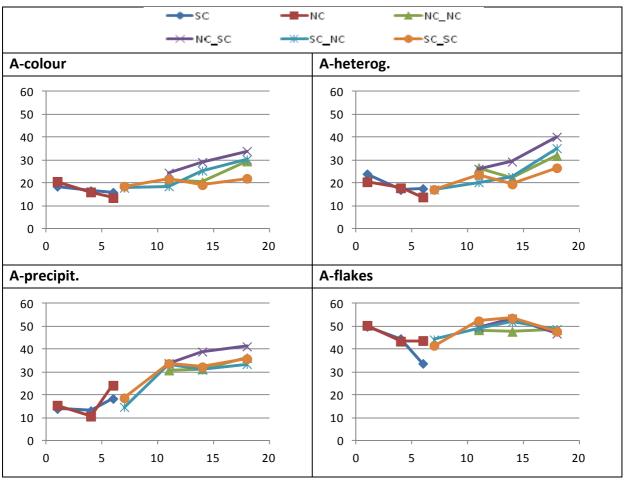


Figure 24. Mean profile scores (scale 0–100, vertical axis) for appearance (A-) attributes for whole cod on days 1, 4 and 6: Superchilled (SC) and Non-superchilled (NC) and cod fillets on days 7, 11, 14 and 18: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC). Horizontal axis – days from catch.

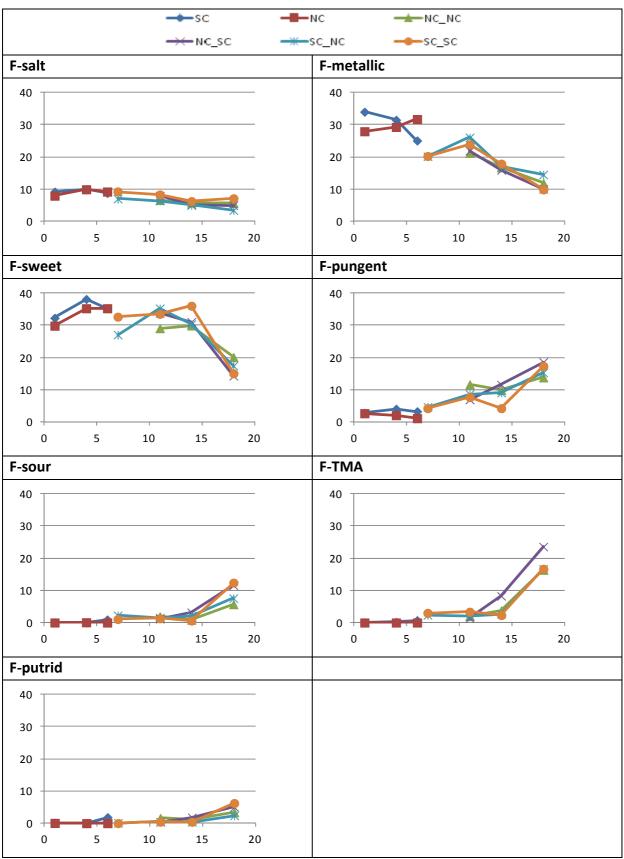


Figure 25. Mean profile scores (scale 0–100, vertical axis) for flavour (F-) attributes for whole cod on days 1, 4 and 6: Superchilled (SC) and Non-superchilled (NC) and cod fillets on days 7, 11, 14 and 18: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC). Horizontal axis – days from catch.

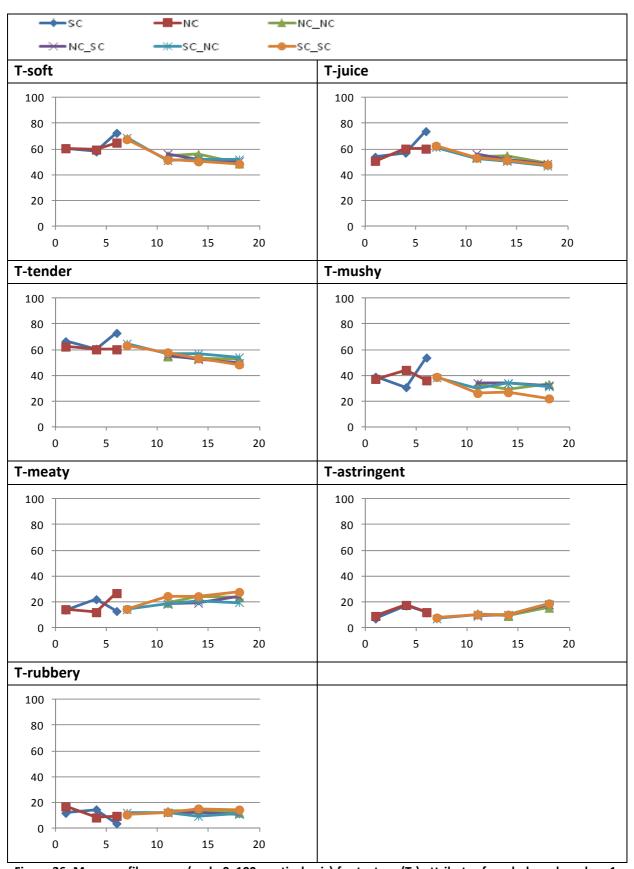


Figure 26. Mean profile scores (scale 0–100, vertical axis) for texture (T-) attributes for whole cod on days 1, 4 and 6: Superchilled (SC) and Non-superchilled (NC) and cod fillets on days 7, 11, 14 and 18: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC). Horizontal axis – days from catch.

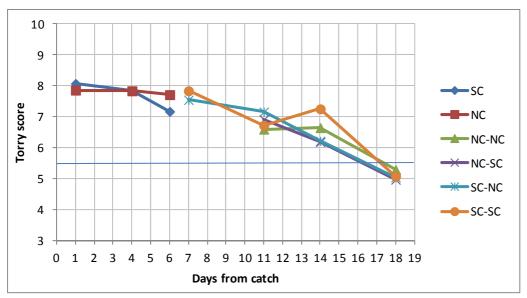


Figure 27. Mean Torry scores for whole cod on days 1, 4 and 6: Superchilled (SC) and non-superchilled (NC) and cod fillets on days 7, 11, 14 and 18: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC).

#### **3B.3 Microbial measurements**

Total viable count was lowest in group NC-NC during the whole storage time but difference between the other three groups were small (Figure 28). Same trends were observed for H<sub>2</sub>S producing bacteria where NC-NC showed the lowest counts (Figure 29). Group SC-NC had in general higher count of H2S producing bacteria apart from day 14 where group SC-SC had the highest count. On day 18 group NC-NC has the lowest count and SC-NC the highest. Difference in count of pseudomonads between groups was not prominent (Figure 30). Group SC-SC had in general the highest count. Group NC-NC had lower count than other groups on day 14 and groups NC-NC and NC-SC had lower count of pseudomonads on day 18 than the other two groups.

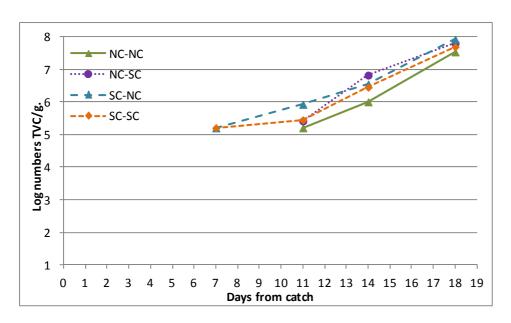


Figure 28. Total viable count per gram on iron agar. Cod fillets: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC), superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC).

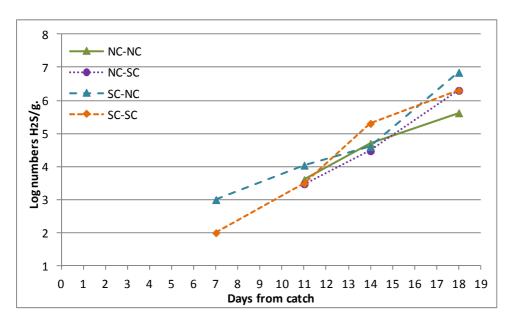


Figure 29. Growth of H<sub>2</sub>S producing bacteria per gram on iron agar. Cod fillets: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC), superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC).

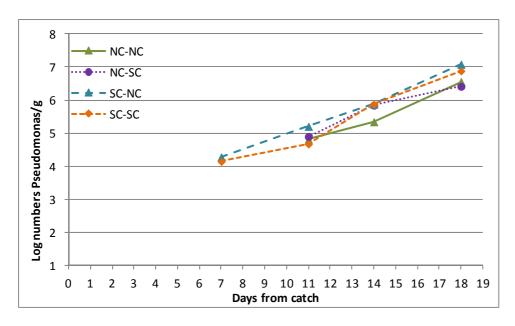


Figure 30. Growth of Pseudomonas bacteria per gram on CFC agar. Cod fillets: Non-superchilled whole, non-superchilled after filleting (NC-NC); non-superchilled whole, superchilled after filleting (NC-SC), superchilled whole, non-superchilled after filleting (SC-NC); superchilled whole, superchilled after filleting (SC-SC).

#### 3B.6 Chemical and physical measurements

#### **3B.6.1 Total Volatile Base Nitrogen**

Levels of TVB-N started to rise after day 14 and on day 18 group SC-SC had the highest level, NC-SC the second highest and groups NC-NC and SC-NC had lower and similar levels (Figure 31).

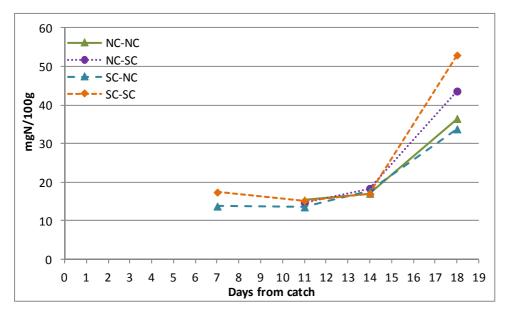


Figure 31. TVB-N values of cod fillets. Non-superchilled whole, non-superchilled after filleting (NC-NC), superchilled after filleting (NC-SC), superchilled whole, non-superchilled after filleting (SC-NC) and superchilled after filleting (SC-SC) on days 7, 11, 14 and 18 from catch.

#### 3B.6.2 pH-measurements

Differences between groups in pH values were minor except on day 14 where group SC-SC had lower pH than other groups (Figure 32). Some difference was also seen on day 7 where SC-NC had slightly higher pH value than SC-SC.

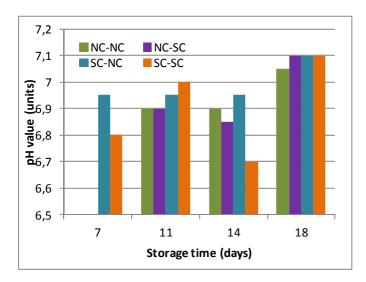


Figure 32. pH values in cod mince of cod fillets. Non-superchilled whole, non-superchilled after filleting (NC-NC); superchilled after filleting (NC-SC); superchilled whole, non-superchilled after filleting (SC-NC) and superchilled after filleting (SC-SC) on days 7, 11, 14 and 18 from catch.

#### 3B.6.3 Water content and water holding capacity

Water content was lowest in group SC-SC during storage time apart from day 18 where it had the highest water content (Figure 33). Group NC-NC also had low water content on days 11 and 18 but higher on day 14. Water content was highest in groups SC-NC but decreased with storage. Group NC-SC was similar to SC-NC.

Water holding capacity (WHC) decreased in group SC-SC but remained stable for other groups during storage.

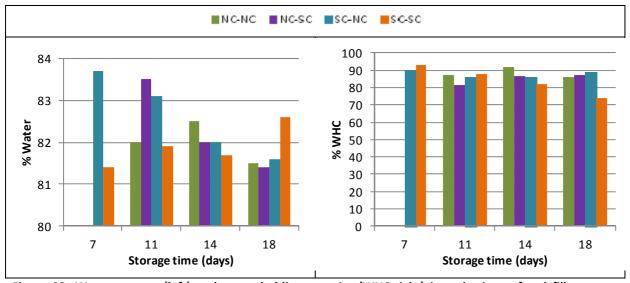


Figure 33. Water content (left) and water holding capacity (WHC-right) in cod mince of cod fillets: non-superchilled whole, non-superchilled after filleting (NC-NC), superchilled after filleting (NC-SC), superchilled whole, non-superchilled after filleting (SC-NC) and superchilled after filleting (SC-SC) on days 7, 11, 14 and 18 from catch.

#### **4 Conclusions**

The results from the sensory evaluation indicate that superchilled processing of whole cod can extend shelf life by two days. However, because normally, the cod is processed no later than 6 days post-catch, the shelf life of whole cod is not as important as the shelf life of fillets. Differences in values of pH, water content and water holding capacity between the superchilled and non-superchilled groups were minor. Similarly, small differences in bacterial growth were seen between non-superchilled and superchilled whole cod during the storage time.

It should be noted that the superchilled storage conditions (mean ambient temperature of  $-1.4 \pm 0.9$  °C and  $-1.3 \pm 0.8$  °C during 18-day storage for the NC and SC groups, respectively) resulted in the same mean product temperature ( $-1.2 \pm 0.2$  °C) in the two groups NC and SC. This made the two groups more similar and might have decreased the effect of the superchilled processing since both groups received the same superchilled storage treatment. Larger differences in chemical and physical properties, bacterial growth and shelf life could be expected in case of chilled storage conditions (0–4 °C) instead of the superchilled conditions in this study.

Differences in Torry scores between the fillet groups were small. Shelf life was estimated between 16 and 18 days which is quite long shelf life for cod fillets. The groups SC-NC and NC-SC had a little shorter shelf life than groups SC-SC and NC-NC, which indicates that the superchilled processing of

either whole cod or fillets was not important. However, the group SC-SC seemed to retain freshness a little longer than other groups. As in case of the whole fish, in general the differences in bacterial numbers, chemical and physical properties between the fillet groups were small. Very similar fillet temperatures between groups resulting from the superchilled storage conditions may be the main reason for the small differences obtained. Thus, another study with more common temperature conditions during transport and storage of fresh fish (chilled but not superchilled) will be conducted.

#### **5** Acknowledgements

This study was a part of the research project "Ofurkældur heill fiskur - fyrir dauðastirðnun" (e. Super-Chilled Round Fish — Pre Rigor), which was funded by AVS R&D Fund of Ministry of Fisheries in Iceland (project no. R 11 062-11). The financing of this work is greatfully acknowledged.

#### **6 References**

Aune EJ (2003). Superchilling of foodstuff, a review. *In:* 21<sup>st</sup> International Congress of Refrigeration, International Institute of Refrigeration. 17-22 August 2003. Washington, DC, USA.

Bjarnason VO (2012). CFD Modelling of Combined Blast and Contact Cooling for Whole Fish. MSc. Thesis, Technical University of Denmark, Department of Mechanical Engineering.

ISO 8586:1993 (1993). Sensory analysis general guidance for the selection, training and monitoring of assessors. Part 1: selected assessors. Geneva, Switzerland: The International Organization for Standardization.

Jorgensen BR, Gibson DM, Huss HH (1988). Microbiological quality and shelf life prediction of chilled fish. International Journal of Food Microbiology 6, 295–307.

Kaale LD, Eikevik TM, Rustad T, Kolsaker K (2011). Superchilling of food: A review. Journal of Food Engineering 107, 141–146.

Magnussen OM, Haugland A, Torstveit Hemmingsen AK, Johansen S, Nordtvedt TS (2008). Advances in superchilling of food – Process characteristics and product quality, Trends in Food Science and Technology 19, 418–424.

Magnússon H, Martinsdóttir E (1995). Storage Quality of Fresh and Frozen-thawed Fish in Ice. Journal of Food Science 60, 273–278.

Magnússon H, Lauzon HL, Sveinsdóttir K, Margeirsson B, Reynisson E, Rúnarsson ÁR, Guðjónsdóttir M, Þórarinsdóttir KA, Arason S, Martinsdóttir E (2009). The effect of different cooling techniques and temperature fluctuations on the storage life of cod fillets (*Gadus morhua*). Tech. report 23-09, Matís, Reykjavík, Iceland. Available at http://www.matis.is/media/matis/utgafa/23-09Differentcoolingtechniques.pdf.

Margeirsson B, Lauzon HL, Sveinsdóttir K, Reynisson E, Magnússon H, Arason S, Martinsdóttir E (2011). Effect of improved design of wholesale EPS fish boxes on thermal insulation and storage life of cod loins – simulation of air and sea transport. Report no. 29-10, Matís, Reykjavík, Iceland. Available at http://www.matis.is/media/matis/utgafa/29-10-Effect-of-improved-design-of-wholesale.pdf

Martinsdóttir E, Magnússon H (2001). Keeping quality of sea-frozen thawed cod fillets on ice. Journal of Food Science 66, 1402–1408.

Martinsdóttir E (2002). Quality management of stored fish. In H.A. Bremner Safety and quality issues in fish processing. p. 360–378, Woodhead Publishing Limited, Cambridge, UK.

Olafsdottir G, Lauzon HL, Martinsdottir E, Oehlenschlauger J, Kristbergsson K (2006). Evaluation of shelf life of superchilled cod (*Gadus morhua*) fillets and the influence of temperature fluctuations during storage on microbial and chemical quality indicators. Journal of Food Science 71(2), S97–S109.

Rha C (1975). Thermal properties of foods, in: Rha, C. (Ed.), Theory, Determination and Control of Physical Properties of Food Materials. Reidel Publishing, Dordrecht, Holland and Boston, USA, pp. 311–355.

Shewan JM, Macintosh RG, Tucker CG, Ehrenberg ASC (1953). The development of a numerical scoring system for the sensory assessment of the spoilage of wet white fish stored in ice. J Sci Food Agric 4:283–298.

Stevik AM, Claussen IC, 2011. Industrial superchilling, a practical approach. Procedia Food Science 1, 1265–1271.

Stone H, Sidel JL (1985). Sensory evaluation practices. Orlando, Fla.: Academic press, Inc. 311p.

Valtýsdóttir KL, Margeirsson B, Arason S, Lauzon HL, Martinsdóttir E (2010). Guidelines for precooling of fresh fish during processing and choice of packaging with respect to temperature control in cold chains. Tech. report 40–10, Matís, Reykjavík, Iceland. Available at http://www.matis.is/media/matis/utgafa/40-10-Guidelines-for-precooling-and-packaging.pdf.

Wang T, Sveinsdóttir K, Magnússon H, Martinsdóttir E (2008). Combined application of modified atmosphere packaging and superchilled storage to extend the shelf-life of fresh cod (Gadus morhua) loins. Journal of Food Science 73, 11-19.

## 7 Appendix

Table 1. Mean values of profiling results for groups NC and SC and p-values for difference between them. Different letters within a line indicate statistically significant difference.

sensory attribute	O-sweet	O-shellfish	O-vanilla	O-potatoes	O-dishcloth	O-TMA	O-sour	O-sulfur	A-colour	A-discol.	A-precipit.	A-flakes
NC_D1	39	34	15	9	2	1	0	0	21	20	15	50
SC_D1	41	32	16	7	1	0	0	0	18	24	14	50
p-value	0,760	0,782	0,824	0,742	0,285	0,392	0,154	-	0,606	0,410	0,638	0,938
NC_D4	43	35	20	6	1	0	0	0	16	18	11	43
SC_D4	45	33	22	9	0	0	0	0	17	17	13	45
p-value	0,843	0,769	0,708	0,524	0,420	0,327	-	-	0,889	0,842	0,540	0,865
NC_D6	39	31	20	6	0	0	0	0	14	14	24	44
SC_D6	37	30	20	7	1	0	0	0	16	18	18	34
p-value	0,832	0,911	1,000	0,692	0,200	-	0,327	0,327	0,389	0,221	0,343	0,228
NC_D11	29	24	22	17	3	2	1	0	20	23	25	44
SC_D11	32	25	19	17	3	3	2	0	21	22	28	50
p-value	0,378	0,701	0,507	0,842	0,728	0,153	0,002 ***	0,010 *	0,844	0,420	0,316	0,019 *
NC-D14	35	23	17	19	15	3	1	0	16	18	28	43
SC-D14	31	23	19	23	15	2	3	0	18	21	31	43
p-value	0,441	0,990	0,761	0,395	0,911	0,645	0,340	0,074 <b>ms</b>	0,371	0,245	0,638	0,936
NC_D18	23	10	7	13	27	15	14	6	19	25	18	50
SC_D18	26	21	13	23	19	5	2	2	15	16	27	53
p-value	0,719	0,225	0,301	0,171	0,283	0,254	0,046 *	0,385	0,383	0,221	0,350	0,773

sensory attribute	F-salt	F-metallic	F-sweet	F-pungent	F-sour	F-TMA	F-putrid	T-soft	T-juicy	T-tender	T-mushy	T-meaty	T-astringer	nt T-rubbery
NC_D1	8	28	30	3	0	0	0	61	51	62	37	14	9	17
SC_D1	9	34	32	3	0	0	0	61	54	67	39	14	7	12
p-value	0,687	0,475	0,756	0,951	0,325	0,325	1,000	1,000	0,595	0,421	0,871	0,930	0,460	0,420
NC_D4	10	29	35	2	0	0	0	60	60	60	44	12	18	8
SC_D4	10	32	38	4	0	0	0	58	57	61	31	22	17	15
p-value	0,983	0,804	0,741	0,329	0,327	0,327		0,811	0,466	0,929	0,052 <b>ms</b>	0,054 <b>ms</b>	0,946	0,266
NC_D6	9	32	35	1	0	0	0	65	60	60	36	27	12	10
SC_D6	9	25	35	3	1	1	2	73	74	73	54	13	12	4
p-value	0,896	0,313	1,000	0,167	0,249	0,327	0,327	0,240	0,012 *	0,038 *	0,030 *	0,015 *	0,978	0,155
NC_D11	7	19	24	6	1	3	0	55	54	57	26	20	15	8
SC_D11	11	26	30	5	4	5	0	54	54	56	25	19	18	9
p-value	0,021 *	0,011 *	0,000 ***	0,402	0,000 ****	0,024 *	0,334	0,689	0,826	0,738	0,725	0,904	0,213	0,633
NC-D14	8	23	30	8	1	3	0	52	46	53	46	19	9	23
SC-D14	9	21	29	10	1	4	0	40	42	43	28	30	16	27
p-value	0,699	0,791	0,893	0,514	0,896	0,868	0,325	0,024 *	0,396	0,034 *	0,003 **	0,071 <b>ms</b>	0,026 *	0,527
NC_D18	7	2	16	15	9	18	19	62	57	67	42	17	12	16
SC_D18	9	11	24	19	5	10	5	36	38	41	16	40	20	36
p-value	0,460	0,045 *	0,412	0,513	0,318	0,293	0,178	0,002 **	0,024 *	0,002 ***	0,028 *	0,007 **	0,240	0,013 *

ms (marginal significance, p = 0.05-0.10); \* (p < 0.05); \*\* (p < 0.01); \*\*\* (p < 0.000)

Table 2. Mean values of profiling results for groups NC-NC, NC-SC, SC-NC and SC-SC and p-values for difference between them. Different letters within a line indicate statistically significant difference.

Group	O-sweet	O-shellfish	O-vanilla	O-potatoes	o-dishcloth	O-TMA	O-sour	O-sulfur	A-colour	A-discol.	A-precipit.	A-flakes
SC_NC-D7	33	23	17	13	2	1	0	0	18	17	15	44
SC_SC-D7	39	24	19	10	1	1	1	0	19	17	19	42
p-value	0,056 <b>ms</b>	0,618	0,371	0,105	0,139	0,699	0,891	-	0,826	1,000	0,093 <b>ms</b>	0,454
NC_NC-D11	37	33	23	22	8	1	1	1	21	27	31	48
NC_SC-D11	36	33	24	24	6	1	1	0	24	26	34	50
SC_NC-D11	39	35	23	25	7	2	2	1	19	20	33	49
SC_SC-D11	35	33	25	23	6	3	1	1	22	24	34	52
p-value	0,455	0,902	0,831	0,750	0,808	0,565	0,676	0,757	0,252	0,155	0,766	0,225
NC_NC-D14	33 b	24 <b>b</b>	19 <b>b</b>	19 <b>b</b>	8	5	2	1 <b>a</b>	20 b	22 b	31	48
NC_SC-D14	32 <b>b</b>	20 b	16 <b>b</b>	30 a	11 <b>a</b>	7 <b>a</b>	3	1	29 a	29 a	39	53
SC_NC-D14	30 b	24	14 <b>b</b>	24	13 a	5	3	0 <b>p</b>	25	23 <b>b</b>	31	52
SC_SC-D14	37 <b>a</b>	30 a	24 a	20 b	3 <b>b</b>	0 <b>p</b>	1	0	19 <b>b</b>	19 <b>b</b>	32	54
p-value	0,002 ***	0,008 ***	0,001 ****	0,003 **	0,023 *	0,065 <b>ms</b>	0,107	0,051 <b>ms</b>	0,029 *	0,006 **	0,078 <b>ms</b>	0,072 <b>ms</b>
NC_NC-D18	23 a	19	16 <b>a</b>	27	21	16	10	3	30	32	36	48
NC_SC-D18	14 <b>b</b>	15 <b>b</b>	10 <b>b</b>	23	21	29	13	4	34 a	40 a	41	47
SC_NC-D18	20	22 <b>a</b>	19 <b>a</b>	28	19	20	11	3	30	35	33	49
SC_SC-D18	18	15	15 a	25	23	26	10	4	22 <b>b</b>	27 <b>b</b>	36	48
p-value	0,058 <b>ms</b>	0,029 *	0,000 ***	0,468	0,931	0,056 <b>ms</b>	0,828	0,543	0,022 *	0,017 *	0,128	0,847

Group	F-salt	F-metallic	F-sweet	F-pungent	F-sour	F-TMA	F-putrid	T-soft	T-juicy	T-tender	T-mushy	T-meaty	T-astringent	T-rubbery
SC_NC-D7	7	20	27 <b>b</b>	5	2	2	0	69	61	65	38	14	7	12
SC_SC-D7	9	20	33 <b>a</b>	4	1	3	0	68	63	63	39	14	8	11
p-value	0,102	0,976	0,028 *	0,753	0,052 <b>ms</b>	0,351	0,337	0,658	0,652	0,534	0,914	0,808	0,228	0,590
NC_NC-D11	7	21	29	12	2	2	2	55	54	55	34 a	19	11	13
NC_SC-D11	8	22	34	7	1	1	0	56	56	56	34	18	9	12
SC_NC-D11	6	26	35	9	2	2	0	51	53	57	30	18	10	12
SC_SC-D11	8	24	34	8	1	3	0	52	53	58	26 b	24	10	12
p-value	0,440	0,081 ms	0,242	0,349	0,911	0,594	0,308	0,354	0,618	0,678	0,034 *	0,181	0,726	0,931
NC_NC-D14	6	17	30	10	1	4 <b>b</b>	1	56	55	54	29	24	9	14
NC_SC-D14	5	16	31	12	3	8 a	2	52	52	52	34	19	10	12
SC_NC-D14	5	17	30	9	2	3 <b>b</b>	0	52	51	57	34	21	10	9
SC_SC-D14	6	18	36	4	1	2 <b>b</b>	0	50	51	53	27	24	10	15
p-value	0,739	0,601	0,077 <b>ms</b>	0,250	0,108	0,011 *	0,325	0,422	0,420	0,539	0,208	0,227	0,798	0,080 ms
NC_NC-D18	6	12	20	14	6	16	3	49	49	52	33	23	15	12
NC_SC-D18	5	10 <b>b</b>	14	19	12	24	5	51	49	50	32	24	18	11
SC_NC-D18	3	15 a	17	15	8	17	2	52	47	54	31	20	18	11
SC_SC-D18	7	10	15	17	12	17	6	49	48	49	22	28	19	14
p-value	0,158	0,053 *	0,419	0,434	0,290	0,267	0,612	0,693	0,872	0,333	0,098 <b>ms</b>	0,428	0,684	0,680

ms (marginal significance, p = 0,05-0,10); \* (p < 0,05); \*\* (p < 0,01); \*\*\* (p < 0,001)