PROJECT REPORT
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Shelf life of chilled radio-frequency and conventionally heated cod and salmon fillets

Radio-Frequency Heating Technology for Minimally Processed Fish Products; RF-Fish
EU project number: QLK1-CT-2001-01788

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Icelandic Fisheries Laboratories Report Summary

Titill / Title	*Shelf life of chilled radio-frequency and conventionally heated cod and salmon fillets*		
Höfundar / Authors	*Ása Þorkelsdóttir, Kolbrún Sveinsdóttir, Kristín A. Þórarinsdóttir, Soffía Vala Tryggvadótti, Emilía Martinsdóttir, Rósa Jónsdóttir, Páll Gunnar Pálsson*		
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Ágrip á íslensku:

Skýrsla þessi er samantekt fimm tilrauna í verkefninu "Radio-Frequency Heating Technology for Minimally Processed Fish Products" árin 2004 og 2005. Fjórar geymsluþolstilraunir voru framkvæmdar á soðnum, vakúmpökkuðum, laxa og þorskbitum sem voru RF (radio frequency)-hitaðir og hitaðir við hefðbundna þrýstisuðu (conventional autoclave) við 75°C og 95°C og geymdir við 3°C. Ferskleiki og áferð voru metin með skynmati og mæld var áferð með tæki, vatnsheldni og þránun. Í fimmtu tilrauninni var soðinn þorskur í lauksósu metinn m.t.t geymsluþols af skynmatshópum í Noregi og á Íslandi.

Samanburður á forsuðuaðferðum og hitunar hitastigi leiddi í ljós svipaða breytingu á ferskleikaeinkennum. Eftir um tvær vikur var farið að greinast aukabragð/óbragð af þorskinum sem var hitaður við 75°C og var ekki munur eftir forhitunaraðferðum. Áferðin var safaríkari, meyrari og flögukenndari í RF hituðum þorski. Eftir tuttugu daga geymslu var súrt bragð og þráabragð farið að greinast í báðum laxahópum. RF laxinn var flögukenndari og meyrari en lax úr hefðbundinni hitun. Ekki náðist lengra geymsluþol með hærra hitastigi (95°C)í forsuðu. Hærra hitastig hafði verri áhrif á áferð og var mjög erfitt að mæla og meta áferðina. Fiskur forhitaður við 75°C var safaríkari og meyrari og flögukenndari þ.e hélt betur lögun sinni en fiskur hitaður við 95°C. Af þessu er dregin sú ályktun að RF hitaður fiskur hafi sambærilegt geymsluþol og hefðbundinn forsoðinn fiskur. Þorskur í lauksósu var af sæmilegum gæðum fyrstu þrjár vikur kæligeymslu, en báðir skynmatshópar voru sammála um að eftir það voru gæðin ekki ásættanleg.

Lykilorð á íslensku: *RF hitun, þorskur, lax, geymsluþol, kæligeymsla, skynmat, áferð, vatnsheldni*

Summary in English:

This report is a part of the EU-project "Radio-Frequency Heating Technology for Minimally Processed Fish Products" and combines five studies carried out in 2004 and 2005. Four quality and shelf life studies were performed on vacuum packed cod and salmon loins that had been RF (radio frequency) and CON (conventional autoclave) heated at 75 and 95°C and stored at 3°C. Freshness and texture was evaluated with sensory evaluation, texture was measured with texture analyser, water holding capacity and rancidity was measured. In the fifth experiment conventionally autoclave heated cod in green onion sauce was evaluated by sensory panels in Norway and Iceland.

Comparison of different heat treatments and temperature during heating showed similar changing in freshness evaluation. After 2 weeks, off-flavour was detected of the cod heated at 75°C and no difference was detected by heat treatment. The texture of RF heated cod was more juicy, tender and flaky. Sour and rancid flavour was detected in both salmon sample groups after 20 days of storage. The RF heated salmon was more flaky and softer compared to the CON heated salmon. Longer shelf life was not obtained by higher temperature (95°C) during pre-heating, and influenced the texture negatively and it became more difficult to evaluate and measure the texture. Fish pre-heated at 75°C was more juicy, tender and flaky and retained its form better compared to fish pre-heated at 95°C. It was concluded that RF heated fish has comparable shelf life to conventionally autoclave heated fish. Cod in green onion sauce was of reasonable quality the first three weeks of chilled storage, but both sensory panels agreed that after that the quality was not acceptable.

English keywords: *Radio-frequency heating, cod, salmon, shelf life, chilled storage, sensory evaluation, texture, water holding capacity*

Shelf life of chilled cod and salmon fillets after radio-frequency and conventional heating

Radio-Frequency Heating Technology for Minimally Processed Fish Products; RF-Fish.

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Comparison of different heat treatments and temperature during heating showed similar changes in freshness attributes. After 2 weeks, off-flavour was detected in the cod heated at 75°C and no difference was found between samples from different heat treatment. The texture of RF heated cod was more juicy, tender and flaky. Sour and rancid flavour was detected in both salmon sample groups after 20 days of storage. The RF heated salmon was more flaky and softer compared to the CON heated salmon. Longer shelf life was not obtained by higher temperature (95°C) during pre-heating, and influenced the texture negatively and it became more difficult to evaluate and measure the texture. Fish pre-heated at 75°C was more juicy, tender and flaky and retained its form better compared to fish pre-heated at 95°C. It was concluded that RF heated fish has comparable shelf life to conventionally autoclave heated fish. Cod in green onion sauce was of reasonable quality the first three weeks of chilled storage, but both sensory panels agreed that thereafter the quality was not acceptable.

English keywords: *Radio-frequency heating, cod, salmon, shelf life, chilled storage, sensory evaluation, texture, water holding capacity*

1. INTRODUCTION

The following experiment was done as a part of the EU project "Radio-Frequency Heating Technology for Minimally Processed Fish Products" (EU project number: QLK1-CT-2001-01788).

Previous reports of this project describe different comparisons of quality measures and products of interest to the project.

In the first part of this project a comparison of quality measurements of fresh and thawed cod fillets was done and showed that sensory analysis, texture measurements and measurement of water holding capacity in addition to microbial counts were all methods that gave valuable information about the quality (Sveinsdottir et al, 2003a).

In the second part of the project four different treatments of cod and salmon fillets were compared; fresh packed, frozen, radio-frequency heated and conventionally heated fillets. In this comparison fresh and frozen/thawed samples were cooked and compared to re-heated samples of radio-frequency heated and conventionally heated fillets. The comparison showed that fresh and frozen/thawed samples received higher freshness scores. Rancid flavour was detected in the re-heated salmon fillets and liquid formation was observed in the re-heated samples (Thorkelsdottir et al, 2004).

The next step in the project was performing shelf life studies of the heated products (cod and salmon), during frozen and chilled storage.
Shelf life studies of pre-heated (radio-frequency and conventionally heated) and raw frozen (individually quick frozen (IQF)) cod and salmon stored at -24°C showed that the products were still acceptable after 9 (cod) and 8 (salmon) months of frozen storage. The pre-heated products differed from the IQF products as the IQF products were flakier and had less water holding capacity compared to pre-heated samples (RF and CON), and the preheated samples were softer (Thorkelsdottir et al, 2005).

Shelf life of fresh fish is short, 9 days for cod fillets stored in ice (Magnússon and Martinsdóttir, 1995) but 15 days for whole cod stored in ice (Martinsdottir, 2001) and 20 days for whole salmon stored in ice (Sveinsdottir et al, 2003b). According to information from fish processing companies (HB-Grandi, Akranes, Iceland), storage time of fresh packed cod fillets is often regarded to be 4-7 days.

Pre-cooked products are well known and pre-cooking may be used to extend shelf life. Processing methods, storage conditions such as temperature and packaging material are important factors in keeping the shelf life as long as possible. However, odour, flavour, appearance and texture will be affected by the heat treatment, resulting in reduced quality despite a prolonged shelf life.
The temperature during heat treatment is known to affect the prolonged shelf life. In a shelf life study of sous vide salmon (González-Fandos et.al, 2005) fish processed at 65°C had shelf life about 21 days when stored at 2°C, which was extended up to 45 days when the product was processed at 90°C.
Expected storage time of pre-cooked cod produced at Fjordkokken are 33 days kept at 0-4°C but 26 days for pre-cooked salmon products kept at 0-4°C.

This report describes the shelf life studies of chilled preheated products. It combines four studies of the year 2004 and 2005. The main objective of the studies was to compare different treatments (Radio-Frequency Heating Technology (RF) used to heat fish loins at two different temperatures and conventionally heated (CON) used to heat fish loins at two different temperatures) of cod and salmon loins packed in vacuum and observe and determine shelf life and quality of those products when stored at 3°C.
The four experiments were the following:

Experiment 3; Shelf life of conventionally (CON) and Radio-Frequency (RF) heated at 75°C cod stored at 3°C.
Experiment 4; Shelf life of conventionally (CON) and Radio-Frequency (RF) heated at 75°C salmon stored at 3°C.
Experiment 5; Shelf life of conventionally (CON) and Radio-Frequency (RF) heated at 95°C cod stored at 3°C.

Experiment 6; Shelf life of conventionally (CON) and Radio-Frequency (RF) heated at 95°C salmon stored at 3°C.

The aim was to estimate maximum shelf life and evaluate quality of those products by using measurements of sensory evaluation, texture measurement with Texture Analyser, Water Holding Capacity (WHC), cook-out %, pH and Thiobarbituric reactive substances (TBARS) content. In addition Formaldehyde (FA) was measured in cod and Peroxide value in salmon.

In addition to the four Experiments mentioned above, a study was carried out to compare the sensory evaluation of two different sensory panels (at the Icelandic Fisheries Laboratories and Fjordkokken) of ready made conventionally heated cod loins in green-onion sauce (see Appendix 1).

2. MATERIAL & METHODS

Sample preparation

Cod (*Gadus morhua*):

Raw material was collected by the fish processing company HB-Grandi (Akranes, Iceland) in May 2004 (Experiment 3) and April 2005 (Experiment 5). After catch, the cod was stored whole in ice for 2-3 days until it was filleted, deskinned and trimmed, loin parts (140±10g) were cut from the fillets, packed in vacuum packs as in previous trials (Sveinsdottir and others, 2003a) and stored at 0-1°C.

The day of packing, the cod samples were packed with ice mats in polystyrene boxes. A part of the raw material was transported to IFL (Icelandic Fisheries Laboratories, Reykjavik, Iceland) for quality check of raw material. Other samples were transported to NORCONSERV (Stavanger, Norway) for conventional heating and to Fraunhofen IVV (Freising, Germany) for RF heating. The samples were heated 4 days after packing and stored at 0-1°C until transported to IFL (Icelandic Fisheries Laboratories, Reykjavik, Iceland), in polystyrene boxes with ice mats. The samples were stored at 3°C until analysed. At the same day as samples were heated in Norway and Germany, raw material was heated at IFL and evaluated as the beginning of the shelf life study.

Salmon *(Salmo salar)*:

Raw material was collected by FK (Fjordkjokken AS, Varhaug, Norway) in August 2004 (Experiment 4) and April 2005 (Experiment 6). Loin parts (140±10g) were cut from the fillets, packed in vacuum pack as in previous trials (Sveinsdottir and others, 2003a) and stored at 0-1°C.

The day of packing, the salmon samples were packed with ice mats in polystyrene boxes. A part of the raw material was transported to IFL for quality check of raw material. Other samples were transported to NORCONSERV for conventional heating and to Fraunhofen IVV for RF-heating. The samples were heated 4 days after packing and stored at 0-1°C until transported to IFL, in polystyrene boxes with ice mats. The samples were stored at 3°C until analysed. At the same day as samples were heated in Norway and Germany, raw material was heated at IFL and evaluated as the beginning of the shelf life study.

Microbial counts

For each sample a 150 g piece of fillet was placed in a filter stomacher bag (from Bagsystem Line, Breveté, France) and 150 g of phosphate buffer added. The bag was placed in the stomacher and blended for 2 min. Then a 1/10 dilution was done: From the filtered section of the bag 22 ml (equals 11 g fish) were pipetted into a new filter bag and 88 g buffer added. Total plate counts were done on Plate Count Agar with 0.5% NaCl by the spread plate technique. First, 1 ml was divided onto 3 plates (1/10). Then tenfold dilution was made as needed. Plates were incubated at 22°C for 3 days. For *Bacillus* spore counts, 10 ml of the 1/10 mixture were heated at 75°C for 30 min. The pour-plate technique was used and the same agar as above. Plates were incubated at 35°C for 2 days.

Sensory evaluation of raw material -cod

Sensory evaluation of raw material was done by the Quality Index Method (QIM) on the whole cod. The QIM scheme lists quality attributes for appearance/texture, eyes, gills, flesh and blood, and descriptions of how they change with storage time. Scores were given for each quality attribute according to the descriptions, ranging from 0-3. Very fresh fish normally receives scores close to 0 with scores increasing with storage time. The scores given for all the quality attributes are added to give the Quality Index, which increases linearly with storage time in ice. The sensory evaluation of each attribute was conducted according to Martinsdottir and others (2001). Two QIM experts from IFL evaluated 10 whole cod from the batch used for Experiment 3 at HB-Grandi.

Sensory evaluation of re-heated and cooked fillets

The sensory evaluation of freshness of cooked cod was done using the Torry scheme giving scores from 10 (very fresh) to 3 (Shewan and others 1953). In addition, the Quantitative Descriptive Analysis (QDA) method (introduced by Stone and Sidel (1985)) was used to assess the cooked samples. The method assumes detailed description of a product, such as odour, flavour, appearance and texture. However, in this project the focus was only on texture for cod and therefore an unstructured scale (0-100 %) was used for a list of words describing texture.

Cooked and re-heated salmon fillets were evaluated by the QDA method (introduced by Stone and Sidel (1985)). Unstructured scale (0-100 %) was used on a list of words of seventeen attributes describing odour, flavour and texture of cooked salmon. Attributes were both positive and negative to evaluate the freshness of samples.

Twelve panellists of the Icelandic Fisheries Laboratories sensory panel participated in the sensory evaluation of the cooked cod. They were all trained according to international standards (ISO 1993); including detection and recognition of tastes and odours, training in the use of scales, and in the development and use of descriptors. The members of the panel were familiar with the Torry and QDA method and experienced in sensory analysis of cod.

The samples were heated at 95-100°C in a pre-warmed oven (Convostherm, Convostar, Germany) with air circulation and steam for 9 minutes in the vacuum packs. Core temperature in samples was 4°C when put into the oven and around 70°C after heating.
Four samples were collect from each loin. The size of each sample was ca. 1-2 cm in width, and 4-6 cm in length. The samples were placed in aluminium boxes (5 cm in width x 8 cm in length x 4 cm in height) and closed with plastic covers before served for the sensory panel. Each sample was coded with a composite of 3 numbers that did not indicate the storage time or any other information. Each panellist evaluated 3 samples in each session and each sample was evaluated in duplicate.
All sample observations were conducted according to international standards (ISO 1988).

Texture measurements

Texture of cooked samples was measured using the Texture Profile Analysis test (TPA). Five cooked loins of cod and salmon were measured from each treatment. The texture analyser used was the TA.XT2i Stable Micro Systems (Stable Micro Systems Ltd., Godalming, England).

The force - time curve was analysed to determine three various texture parameters: *Hardness*: The maximum force (N) at certain adjusted deformation. *Cohesiveness*:

Amount (%) of displacement before the sample brakes (strength of inner bonds). *Resilience*: The capability (%) of a strained body to recover after deformation caused by compressive stress.

The SMS probe and setting for the TPA test were: Aluminium Compression plate, 100 diameters (P/100). Pre test speed 2,0 mm/s; speed in sample 0,8 mm/s. Strain (distance) 80%. Load Cell Capacity (kg) 25. During a TPA test the sample was compressed two times in a reciprocating motion that was supposed to imitate the action of the jaw.

The samples were cooked in the vacuum packs in a steam oven (95-100°C) for 9 minutes and put on ice, and stored in 2°C refrigerator for minimum 2 hours before measured. Two to three cm of the top (neck part) of each fish sample (loin) were removed, then three 2.5-cm slices were cut across the fillet and each slice cut into 2.5-cm cube (sample size 2.5 * 2.5 cm). All fish pieces and prepared samples were stored on plastic film on ice until measured. The reported TPA force for each fillet (loin) was the average value of 3-4 measurements.

Cook-out

Evaluation of cook-out was performed by steam cooking/heating the vacuum packed loins (n=3) at 95-100°C for 10 min in a Convostar oven (Convotherm, Elektrogeräte GmbH, Eglfing, Germany). Core temperature in samples was in the range of 2 to 4°C when put into the oven.

The loins were cooled on ice for 15 min prior to weighting. The total weight of the vacuum packed loins was recorded, then the package was cut open and the cooked-out liquid pored away. Then weight of the fish and packing material was recorded and finally only the packaging material was weighted. The values obtained were used to calculate the cook-out, which was expressed as percent of the weight lost due to cooking.

$$\text{Cook-out \%} = 100 \times \frac{\textit{Weight of sample in packaging} - (\textit{Weight of drained sample} + \textit{packing material})}{\textit{Weight of sample} - \textit{packing material}}$$

Water holding capacity (WHC)

The analysis of WHC was based on method described by Børresen (1980) but was modified by reducing the speed from 1500 g`s to 500x g`s. Cooked samples (n=3) were stirred with a spatula to homogenise the sample. Approximately 2 g of the muscle were weighted accurately into a test tube with known weight and centrifuged (SS-34 rotor; Sorvall RC-5B, Du Pont, Delaware, USA) at 530g for 5 min; with temperature maintained at 2 to 5 °C. Two parallels were used for each sample. After centrifugation, the total weight of each test tube and sample was recorded and used to calculate sample weight after centrifugation. WHC was calculated as percentage remaining water of initial water in sample:

$WHC\ (\%) = (v_1-\Delta r)/(100-\Delta r)* 100\%$

v_1 = % water in sample before centrifugation

v_1 = (Weight before drying-Weight after drying) / (Weight before drying) * 100%

Δr= Weight before centrifugation - Weight after centrifugation / (Weight before centrifugation) * 100%

Water content, fat content and pH

Water content (g/100g) was calculated as the loss in weight, after drying at 105 °C for 4 h (ISO 1983). Fat content was determined by the AOCS Soxhlet method Ba 3-38 (AOCS, 1998) using petroleum ether (Bp. 40-60 °C) for extraction. The pH was measured before WHC-analysis by inserting a combination electrode (SE 104, Mettler Toledo GmbH, Greifensee, Switzerland) directly into the samples. The electrode was connected to a portable pH meter (Portamess 913 pH, Knick, Berlin, Germany).

Peroxide value

Extraction of lipids was carried out by chloroform/methanol extraction system based on the method of by Bligh and Dyer (1959) with some modifications (Hanson and Olley, 1963) and with butylated hydroxytoluene (BHT) admixed into all solvents (50-100 mg/L). The following determinations on the lipid fraction were performed after evaporation (Büchi, Switzerland) at 37 °C under vacuum. Peroxide value (meq/kg lipid)

of the extracted lipids was measured by iodometric titration according to AOAC official method 965.33 (AOAC, 1990).

Thiobarbituric reactive substances (TBARS)

TBARS were determined by a modified version (Sørensen & Jørgensen 1996) of the extraction method described by Vyncke (1970, 1975) with few modifications. The sample size was reduced to 15 g and homogenized with 30 mL of 7.5% trichloroacetic acid solution containing 0.1% of both propyl gallate and EDTA. The absorbance of samples and standards were measured at 530 nm. TBARS, expressed as µmol malondialdehyde per kilogram of sample (µmol MDA/kg), was calculated using malondialdehyd-bis-(diethyl acetate) as standard.

Formaldehyde (FA) content

Samples were prepared with addition of phosphoric acid and distillation of formaldehyde and then react with cromotropicacid. Absorbance were measured at 530 nm. (Z.Anal. Chem. 1937).

Data analysis

Statistical analysis was performed by Microsoft Excel 8.00 (Microsoft Inc, Redmond, USA) and NCSS 2000 (NCSS, Utah, USA). Student's t-test, ANOVA and Duncan's test were performed to analyse if the samples were statistically different. Multivariate comparison of the different attributes measured was carried out in the statistical programme Unscrambler ®, Version 6.1 (CAMO, Trondheim, Norway), with principal component analysis (PCA). Before the analysis, variables were scaled. Each element in the matrix was multiplied with the inverse of the standard deviation of the corresponding variable if the variables had different ranges. By doing this, each variable has the same variance. The significance level was $p < 0.05$.

3. RESULTS AND DISCUSSION

The following chapter describes the results of experiments 3, 4, 5 and 6.

EXPERIMENT 3; SHELF LIFE OF CONVENTIONALLY (CON) AND RADIO-FREQUENCY (RF) HEATED AT 75°C COD STORED AT 3°C

Microbial counts

Total viable counts are shown in table 1. The values were within the limits of good quality of raw material defined in the project (Palsson et al 2005).

Table 1. Microbial counts (Total plate counts at 22°C) cfu/g.

Days after precooking	Sample	TVC
	Raw material	15300
day 6	CON	<10
day 6	RF	140
day 14	CON	EST<10
day 14	RF	EST 40
day 18	CON	EST<10
day 18	RF	EST 40

Sensory evaluation of raw material

At the processing day average QI for 10 evaluated cod was 0.9 and within the limits of acceptable freshness in this project i.e. QI<4. Temperature in evaluated fish was -0.2°C-0.9°C and after packing of fish loin parts the temperature was -0.8°C.

Sensory evaluation

At the same day as the heating process was performed in Norway and Germany, 3 days after packing of the raw material, samples were cooked at IFL and evaluated. The samples are named and called storage day 0 in the shelf life experiment. RF and CON

heated samples were reheated and evaluated after 6, 14 and 18 days at 3°C storage. The results are shown in table 2 and Figure 1 and Figure 2.

Table 2. Average sensory scores of cod samples as evaluated by the sensory panel. Different superscripted letters indicate difference (p<0.05).

Sample Days from cooking	Freshness Torry score	Flakes	Softness (firm/soft)	Juiciness (dry/juicy)	Tenderness (tough/tender)
Raw material-D0	9,0[a]	59	56	60	56
RF- D6	7,8[b]	55	58	48	57
CON - D6	7,5[b]	47[a]	46	43	44
RF - D14	5,7[c]	57	59	52	59
CON - D14	5,4[c]	57	50	47	46
RF- D18	5,5[c]	63[b]	59	48	58
CON- D18	5,6[c]	57	44	47	43

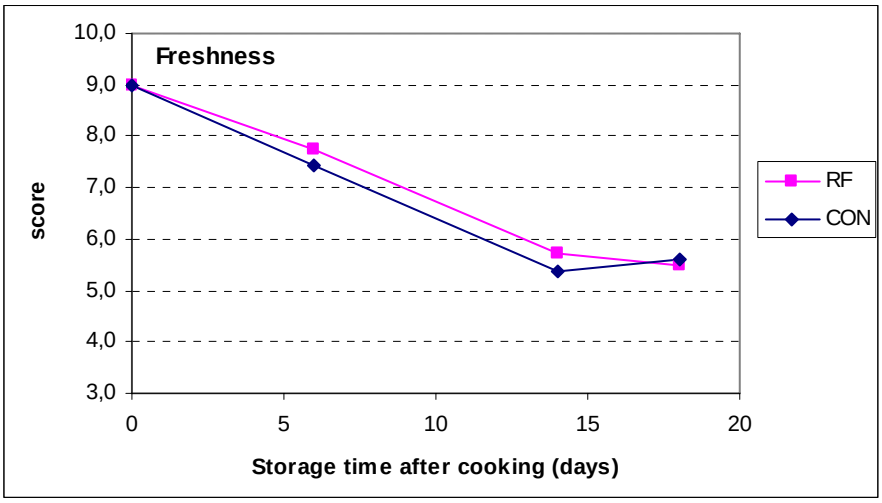

Figure 1. Average Torry scores of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

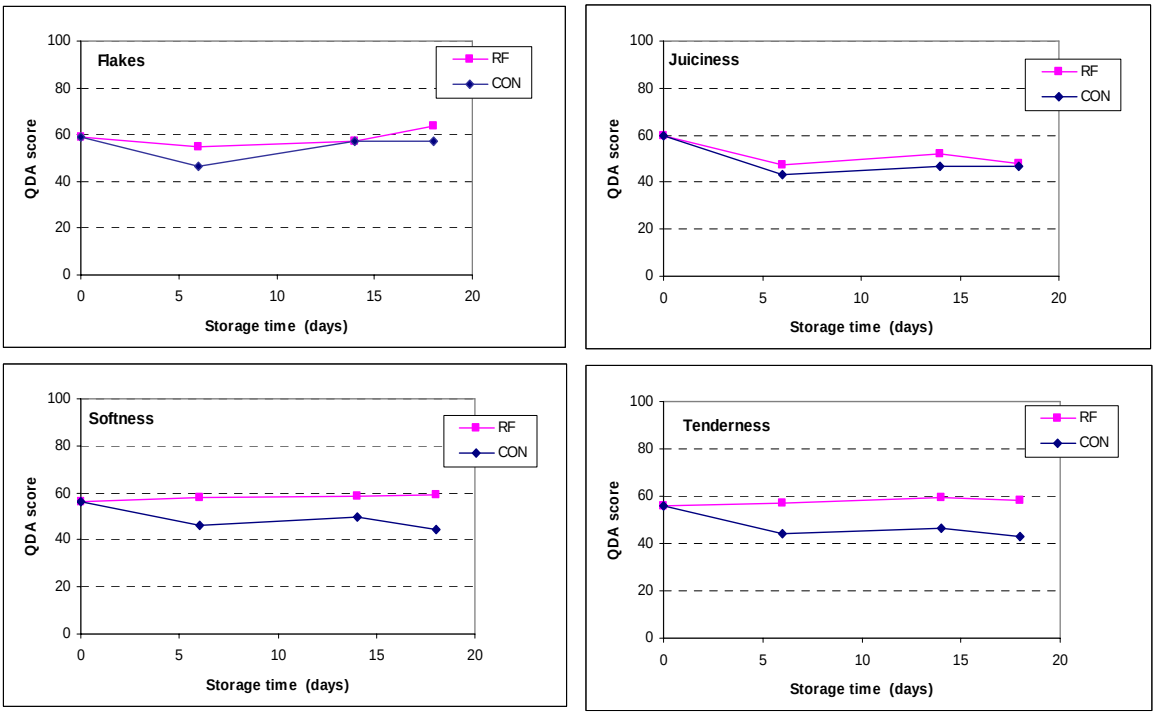

Figure 2. Average QDA (texture) scores of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Freshness decreased significantly with the storage time until storage day 14. Then the average freshness scores were around 5.5 which is considered as the limits for shelf life as evaluated with the Torry freshness score sheet by IFL. Samples were evaluated similar at day 18. At each sampling day RF and CON samples were not different with regard to freshness. RF samples were slightly softer and more tender than CON, though not statistically significant. Flakiness, juiciness, softness and tenderness were not affected by storage time.

The loss of freshness was faster than expected at the beginning of the trial and comments from the sensory panel indicated that some off flavour affected the freshness analysis. This off flavour might have been warmed over flavour (WOF), which can occur in re-heated meat and fish after short chilled storage. The formation of WOF has been studied for meat and meat products, but very little for fish. WOF is described as spoilage odour which can remind of cardboard, paint or rancidity (Vega and Brewer 1994; Love 1988).

Meat rich in poly unsaturated fat acids (PUFA) is more likely to contain WOF according to Cross et al (1987) who showed that the speed of WOF formation in the following products was correlated to their PUFA content: fish > chicken > pork > beef > lamb.
Some research have shown that cooking prevents enzymatic activity resulting in prolonged shelf life compared to the shelf life of fresh fish (Refsgaard et al 1998; Refsgaard et al 2000). However, more research indicate enhanced oxidation after cooking because of ruptured cell membranes, denatured membrane proteins and increased accessibility for e.g. oxygen, iron and other rancidity catalysts to fat acids (Mielche and Bertelsen 1994).
Research on heating or cooking of fish has shown different effects on rancidity (Hardy 1980; Undeland et al 1998). Undeland et al (1998) showed that pre-cooking inactivated enzymes that catalysed rancidity in minced herring. However, the activity of rancidity blockers was reduced and in general heat stimulating catalysing of rancidity increased.

It could not be overruled that the off flavour might also have been from the packaging material used for the cod, the plastic bag each loin was packed and cooked in. The off-flavour could have been due to edge overheating and too much thermal stress on packaging film. Available information on the film used was described in a previous report by Sveinsdottir et al (2003a) and the sealing range for the plastic material should have been in the range of 115-155°C.

Due to this, some analysis of oxidation was added and packing material was changed in the forthcoming experiments.

Instrumental texture measurements

The measured average values for hardness of cooked cod are shown in Figure 3, the values for cohesiveness and resilience of the same samples are shown in Figure 4 and Figure 5 respectively. RF and CON samples were different on the sixth day of storage and the fourteenth day with regard to hardness, but no differences were found for cohesiveness. Resilience was higher in CON heated samples compared with RF heated samples but the difference was only significant at the fourteenth day of storage.

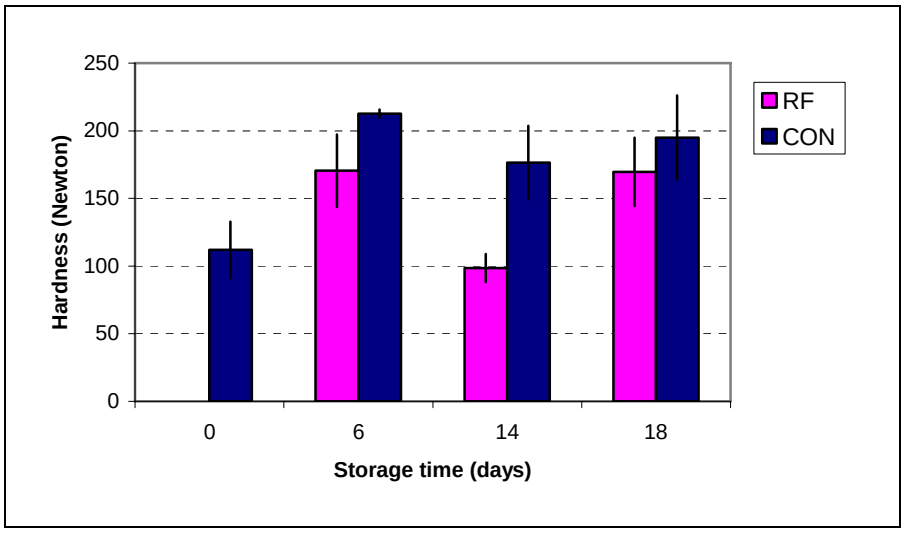

Figure 3. Hardness measurement of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

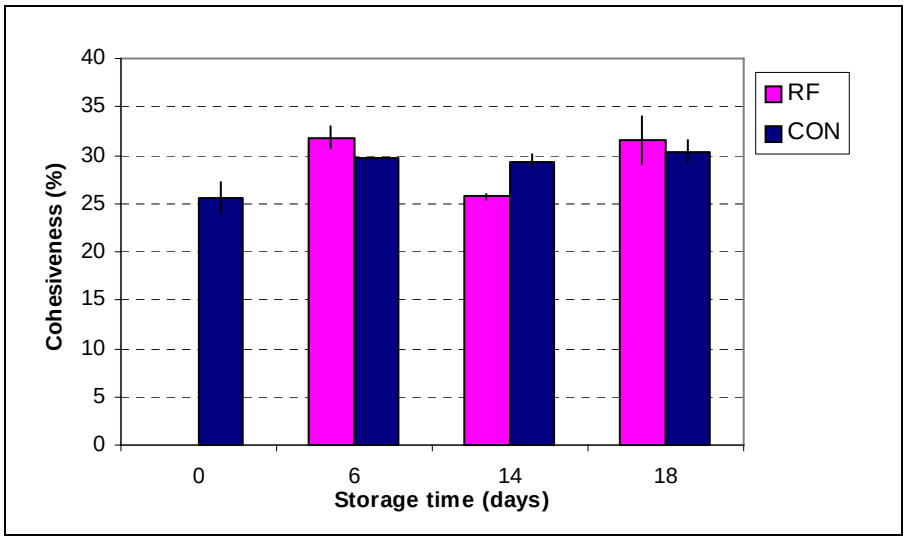

Figure 4. Cohesiveness of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. Calculated values with standard deviation from the time–force curve of TPA measurement. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

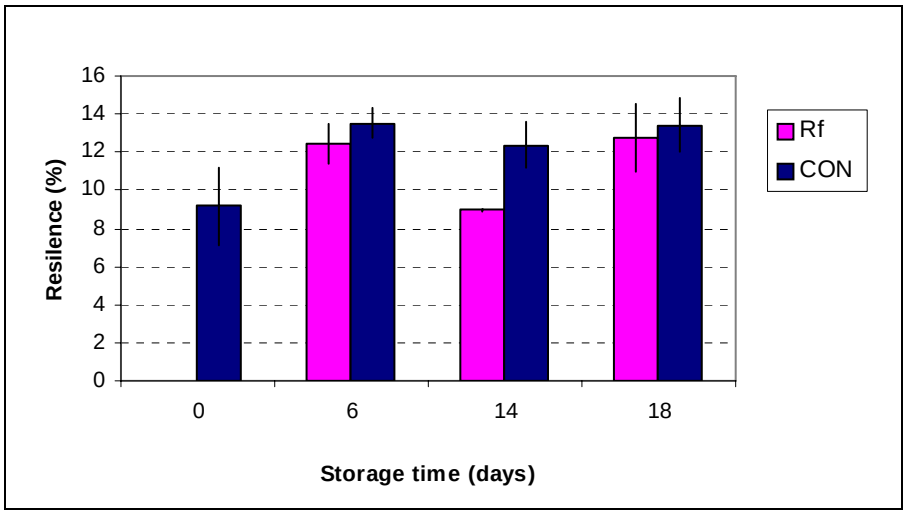

Figure 5. Resilience of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. Calculated resilience (%) values with standard deviation from the time–force curve of TPA measurement. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Water content and pH

The water content of fresh heated cod was 76.9 ± 0.6% and pH 6.58 ± 0.01. The water content of the CON samples was lower (74.2-75.1%) than of RF samples (76.6 – 77.4%) but effects of storage time were not significant. Effects of heat treatment and time were similar for pH, it was lower (6.48-6.64) in CON than in RF heated samples (6.64-6.74) but the variation with time was low (Figure 6).

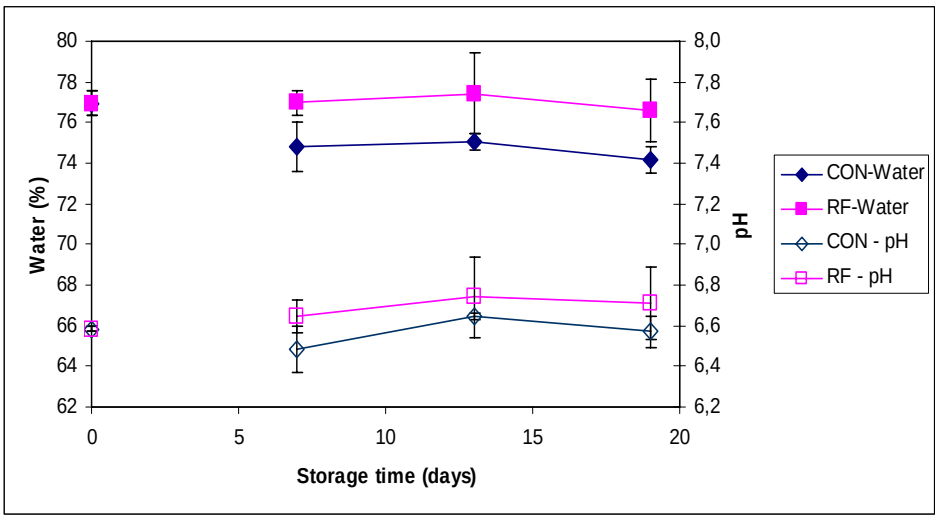

Figure 6. Water content (%) and of cooked/re-heated cod (n=3); RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Cook-out and water holding capacity (WHC)

The cook-out (%) of the fresh heated cod was 24.3 ± 2.6% and the WHC 68.9 ± 3.0%. After 7-18 days of storage, the cook-out of the RF samples was similar (24.3 – 26.2%) but higher in CON samples (33.3 – 35.0%). The difference in WHC between CON (75.3 – 76.0%) and RF samples (73.0 – 73.7%) was not significant, although slightly higher in CON. The effects of storage time did not affect cook-out (%) or the WHC (Figure 7).

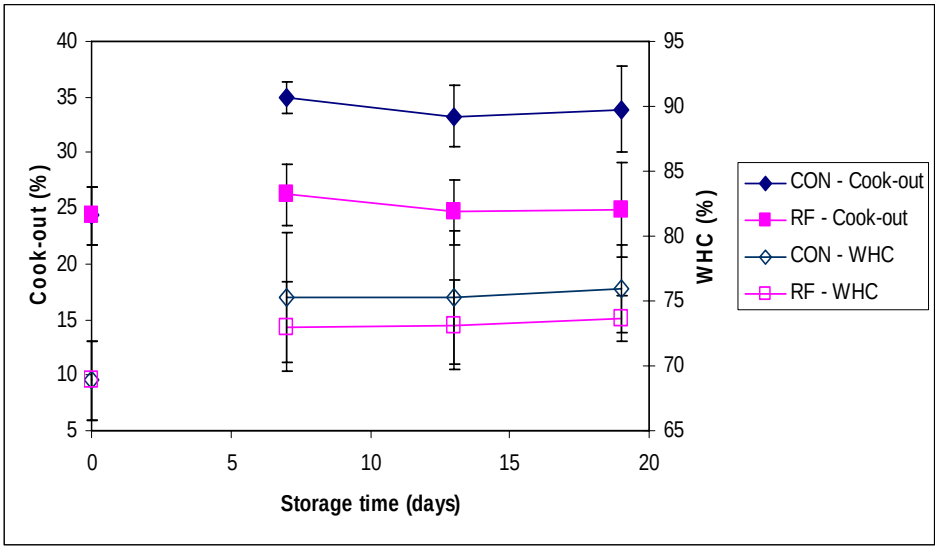

Figure 7. Cook out and WHC (%) of cooked/re-heated cod (n=3); RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Thiobarbituric reactive substances (TBARS)

TBARS was analysed after 21 days of chilled storage in pooled samples. For comparison, fresh untreated (not from the same batch) sample was cooked and analysed. The TBARS in the fresh samples was 1.3 µmol/kg which indicated low content of secondary oxidation products. It was similar in CON and RF-heated samples 11.5 and 10.9 µmol/kg, respectively.

EXPERIMENT 4; SHELF LIFE OF CONVENTIONALLY (CON) AND RADIO-FREQUENCY (RF) HEATED AT 75°C SALMON STORED AT 3°C

Microbial counts

Total viable counts are shown in table 3. The TVC of the raw material was within the limits of good quality of raw material defined in the project (Palsson et al 2005). The TVC of the precooked products was very low, indicating that the heat treatments were sufficient.

Table 3. Microbial counts (Total plate counts at 22°C) cfu/g.

Days after precooking	Sample	TVC
	Raw material	16000
day 7	CON	EST<10
day 7	RF	EST 10
day 14	CON	EST 70
day 14	RF	EST<10
day 20	CON	100
day 20	RF	EST <10

Sensory evaluation

Samples were evaluated with sensory evaluation after seven, fourteen and twenty days of storage at 3°C. The results are shown in table 4 and Figure 8.

Table 4. Average sensory scores of salmon samples as evaluated by the sensory panel. Different superscripted letters indicate difference ($p<0.05$). CON; conventionally heated, RF; radio-frequency heated, RM; raw material heated at IFL, D=days in storage at 3°C, o = odour, f = flavour.

Sample	characteristic (o)	seaweed-sea (o)	liver-oil (o)	earthy (o)	sour (o)	rancid (o)	flakes	softness	juiciness	tenderness	characteristic (f)	metal (f)	oil (f)	sweet (f)	earthy (f)	sour (f)	rancid (f)
RM	61	38	26	27	6^{b}	4^{b}	52	61	62	65	62	34	36	26	37	7^{b}	5^{b}
RF D07	53	29	21	29	7^{b}	16^{b}	50	55	53	62	61	31	39	22	27	4^{b}	13^{b}
CON D07	47	21	22	32	6^{b}	13^{b}	44	61	58	65	58	29	36	23	27	3^{b}	12^{b}
RF D14	46	29	31	35	11	10^{b}	46	49	48	60	50	33	37	32	30	14	16^{b}
CON D14	46	26	25	31	4^{b}	9^{b}	53	64	52	67	54	27	37	15	38	5^{b}	17^{b}
RF D20	48	27	23	35	19^{a}	33^{a}	57	54	51	57	49	36	35	25	35	19^{a}	32^{a}
CON D20	51	20	29	36	10	11b	52	63	56	62	50	17	43	17	43	12	19^{b}

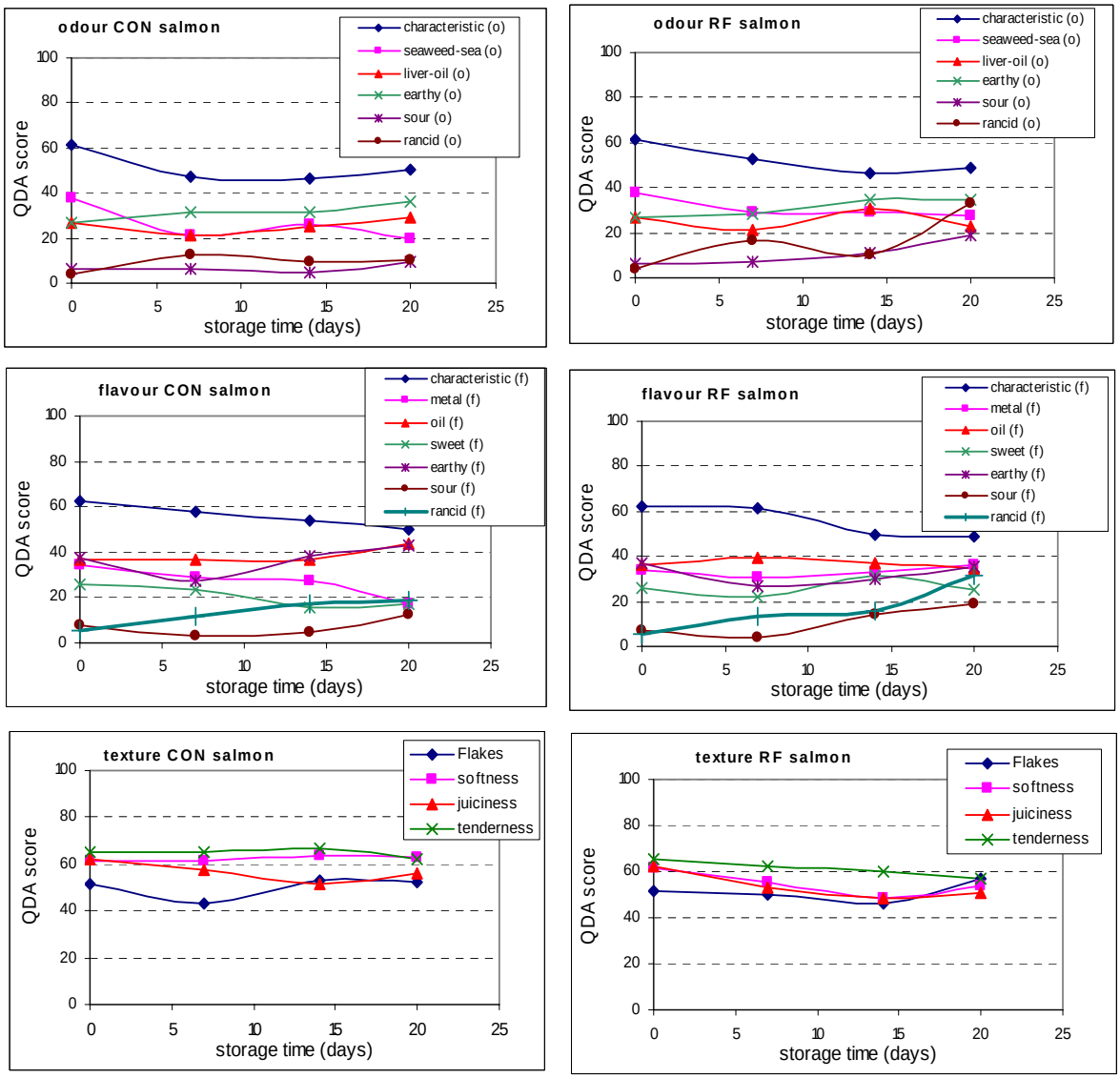

Figure 8. Average QDA (texture) scores of cooked/re-heated salmon; RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 0, fresh salmon was analysed after heating at IFL. Storage at 3°C

There was no significant difference between RF and CON samples first fourteen days of storage. Higher scores were observed for rancid odour and flavour in reheated samples compared to the raw material cooked at IFL, similar as in previous experiments 2003 (Thorkelsdottir et al, 2004). After twenty days of storage RF heated samples had significantly higher intensity of sour and rancid odour and flavour compared to samples at other sampling days including CON heated samples at day 20. Texture did not change significantly during the storage.

Instrumental texture measurement

Measured average values for hardness, cohesiveness and resilience are shown in Figure 9, 10 and 11 respectively. All these parameters were higher in the reheated salmon samples than measured in the raw material, samples cooked at IFL. On the seventh day of storage hardness and resilience were higher in RF heated samples than CON heated samples. At the end of the shelf life there was samples were not significantly different in the measured texture parameters.

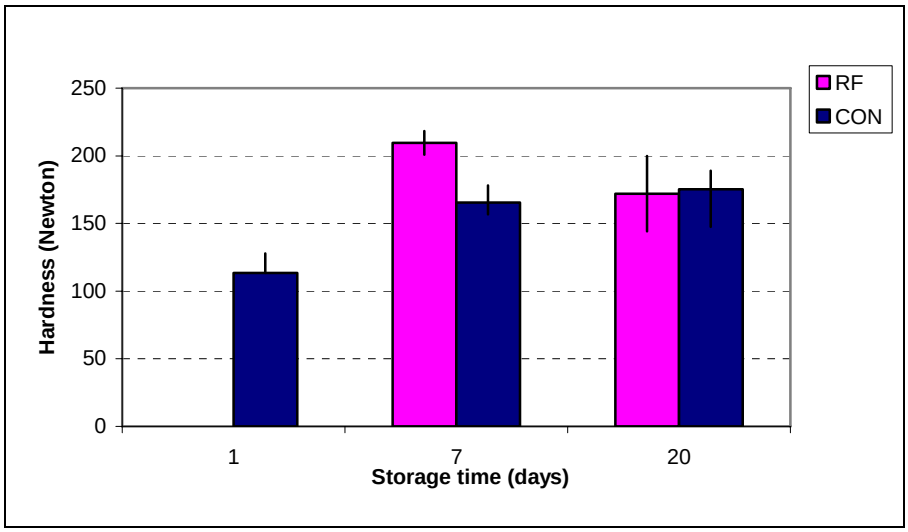

Figure 9. Hardness measurement of cooked/re-heated salmon; RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 1 fresh cod was analysed after heating at IFL. Storage at 3°C.

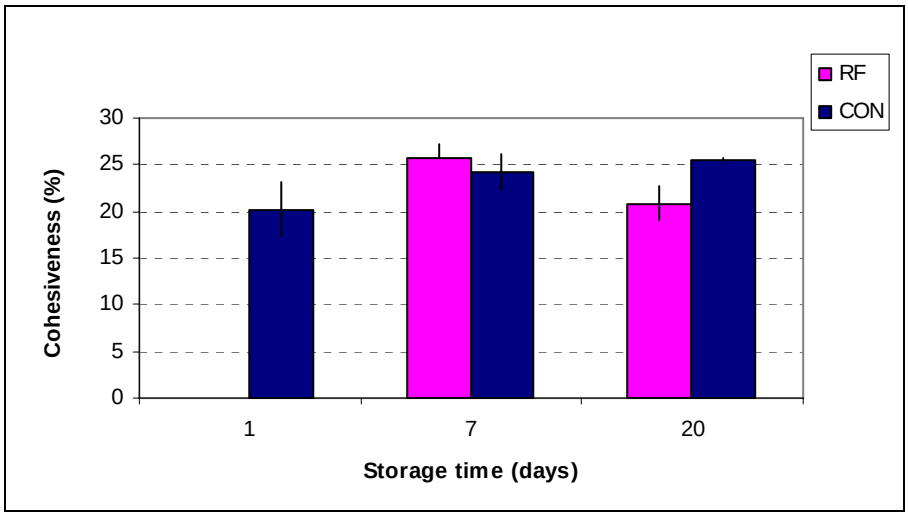

Figure 10. Cohesiveness of cooked/re-heated salmon; RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 1, fresh cod was analysed after heating at IFL. Storage at 3°C.

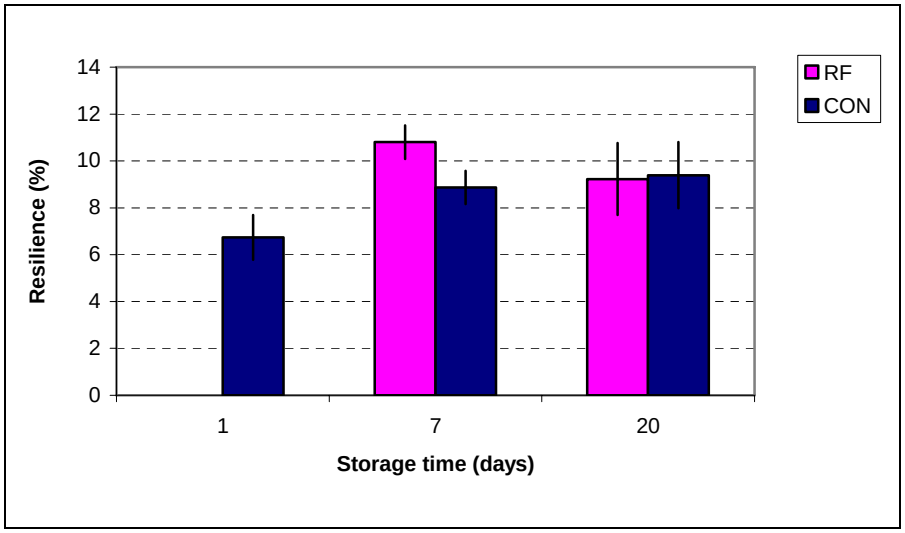

Figure 11. Resilience of cooked/re-heated salmon; RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 1, fresh cod was analysed after heating at IFL. Storage at 3°C.

Water content, fat content and pH

The water content of CON and RF-heated salmon samples (63.6 and 65.5%) on day 7 was higher than in fresh heated samples (60.6%). The water content was higher in the

RF-heated samples than in CON-heated samples during the whole period. Effects of time during chilled storage of heated samples were only significant between day 7 and 20.

Changes in fat content were in converse to water content as expected and it was higher in CON than RF-heated samples. Values obtained were lower than in the fresh heated samples but remained similar during storage from 7 to 20 days (Figure 12).

The pH in fresh heated samples was 6.30 ± 0.02. It was higher in RF-heated samples than in CON-heated samples, but the difference was only about 0.1. The variation with time was not significant (Table 5).

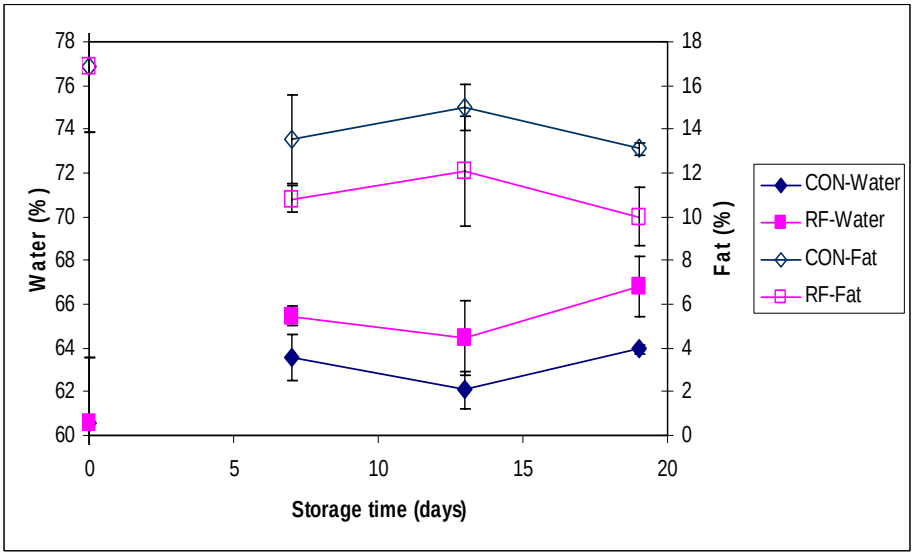

Figure 12. Water and fat content (%) of cooked/re-heated salmon (n=3); RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Table 5. pH of precooked samples (n=3) which were reheated after 7-20 days at 3°C (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh salmon was analysed after heating at IFL

Storage (days)	7	13	20
CON	6,32 ± 0.01	6,28 ± 0.04	6,27± 0.01
RF	6,38 ± 0.05	6,33 ± 0.02	6,40 ± 0.06

Cook-out and water holding capacity (WHC)

Cook-out (%) was lower in the fresh salmon (9.7 ± 1.2%) than in CON (17.1-17.6%) and RF-heated samples after 7 to 20 days of chilled storage (12.4-17.8%). It was similar in CON-heated samples during the whole storage period but decreased slightly in RF-heated samples (Figure 13). On the latest sampling day, the difference between RF and CON heated samples was significant.

WHC (%) was higher in the RF-heated (64.6-68.5%) samples than in CON (58.3-62.7%) samples. Higher cooking loss in the RF and CON samples may partly explain higher WHC than in the fresh heated salmon (50.6 ± 7.4%) since more of the loosely bound water was lost during cooking and therefore the remaining water was likely to be more firmly bound in the muscle.

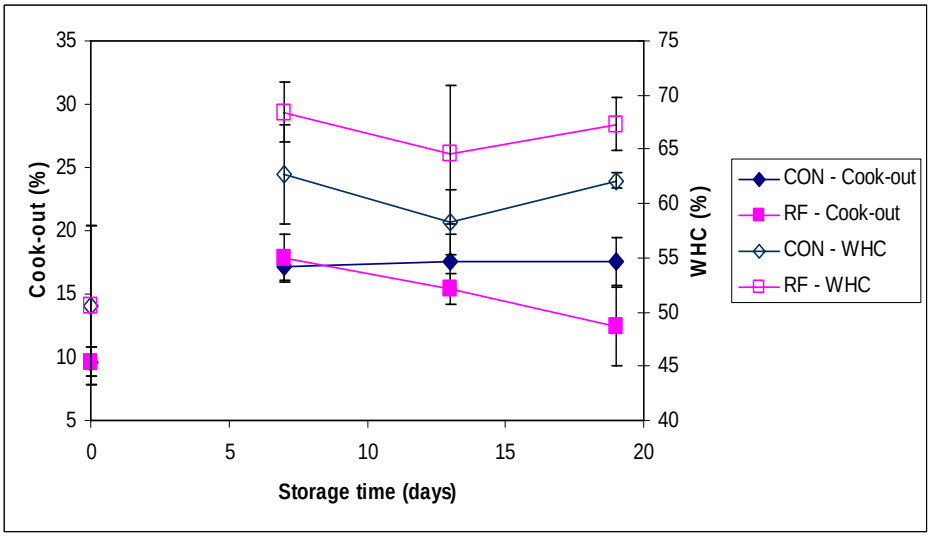

Figure 13. Cook out and WHC of cooked/re-heated salmon (n=3); RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C.

Peroxide value (PV) and thiobarbituric reactive substances (TBARS)

The PV in fresh heated salmon was 8.8 meq/kg but slightly higher in CON and RF samples on day 7, of 9.5 meq/kg and 9.4 meq/kg respectively. From day 7 to 14 it increased in RF-samples but decreased rapidly from day 14 to 20 in both CON and RF-

heated samples, to 2.9 and 5 meq/kg respectively (Figure 14). The values for the fresh salmon indicated that oxidation had started in the raw material. As an example for fresh salmon it can be mentioned that Fagan et al (1998) found that PV in fresh salmon was 1.6 meq/kg.

The TBARS in fresh salmon after cooking was 42.6 μmol/kg. After 7 days of chilled storage, the TBARS of the CON-heated samples (37.0 μmol/kg) was higher than of RF-heated salmon (27.7 μmol/kg). TBARS decreased continuously in CON during storage and the final value after 20 days was 30.1 μmol/kg. The TBARS in RF heated samples decreased only slightly from day 7 to day 13 (27.7 and 26.1 μmol/kg respectively). After that TBARS increased to a final value of 30.3 μmol/kg which was similar to the TBARS in the CON heated sample on day 20 (Figure 14).

Peroxide values have been used to estimate the initial products of lipid oxidation and TBARS to evaluate secondary products. It is known that PV may decrease after certain time and that the secondary products are not end-products of lipid oxidation and may react further with other components of the fish (Auburg 1993) which may lead to decreasing TBARS. What makes the interpretation of PV and TBARS difficult, is that these factors in our trial were analysed in cooked samples and may as such only function as reference values for later studies since little experience has been gained with cooked fish.

It is known that the extent of lipid oxidation may increase after cooking. The process probably disrupts the muscle membrane system, thereby exposing the lipid components to oxygen and other reaction catalysts such as iron (Hardy 1980, Flick et al. 1992, Mielche & Bertelsen 1994, Undeland et al. 1998). However, the way of cooking affects changes in the lipids during cooking and some studies have shown that oxidation products remained similar or changed only slightly (Al-saghir et al 2004, Regulska-IIow and IIow 2002). It has also been hypothesed that high levels of natural antioxidants in red flesh fishes of the Salmonidae family may prevent degradation of polyunsaturated fatty acids during heat treatment (Gladyshev et al 2005).

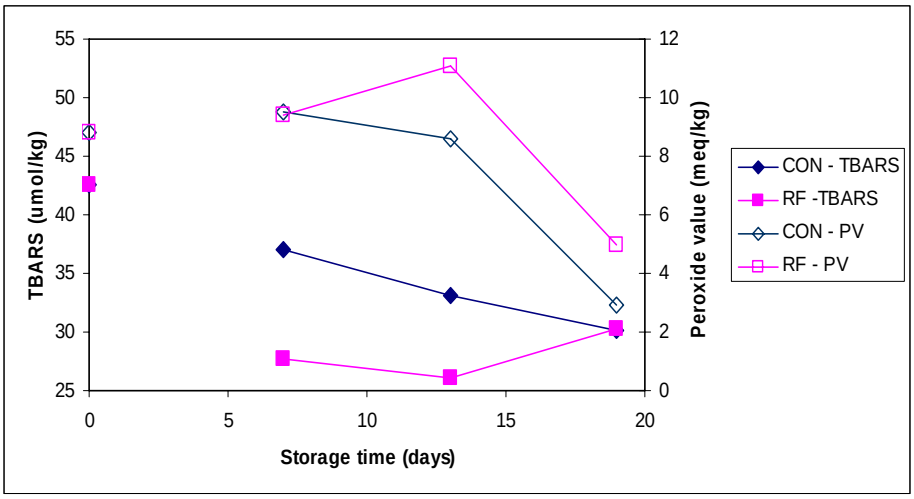

Figure 14. TBARS and PV of cooked/re-heated salmon (pooled samples (n=3); RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon. On day 0, fresh cod was analysed only after heating at IFL. Storage at 3°C.

EXPERIMENT 5; SHELF LIFE OF CONVENTIONALLY (CON) AND RADIO-FREQUENCY (RF) HEATED AT 95°C COD STORED AT 3°C

Because of failure in packaging of cod samples, which was detected during RF heating process of the samples, the produced amount of samples was reduced. Because of this it was not possible to carry out all planned measurements.

Microbial counts

Total viable counts are shown in table 6. The values in raw material were within the limits of good quality, defined in the project. At the 21st storage day, TVC was 550 in RF heated samples. In other heated samples TVC was estimated 30 or lower.

Table 6. Microbial counts (Total plate counts at 22°C) cfu/g.

Days after precooking	Cod sample	TVC
	Raw material	11000
day 7	CON	est 20
day 7	RF	est<10
day 21	CON	est 30
day 21	RF	550

Sensory evaluation of re-heated and cooked fillets

Fresh cod (raw material) was cooked at IFL and evaluated as storage day 0 in the shelf life study. RF and CON heated samples were reheated and evaluated after 7 and 21 days after storage at 3°C. The results are shown in table 7 and Figure 15 and Figure 16.

Table 7. Average sensory scores of cod samples as evaluated by the sensory panel. Different superscripted letters indicate difference ($p<0.05$). RM (fresh sample cooked at IFL), RF (Radio-Frequency heated, CON (Conventionally heated).

Sample	Freshness Torry score	Flakes	Softness (firm/ soft)	Juiciness (dry/ juicy)	Tenderness (tough/ tender)
RM95	$8,6^{a}$	58^{a}	56^{ac}	51^{ac}	53^{b}
CON 95 d7	$6,9^{b}$	31^{b}	67^{ab}	44	51^{b}
RF 95 d7	$7,2^{b}$	55^{a}	74^{b}	49^{c}	70^{a}
CON 95 D21	$5,4^{c}$	45	46^{c}	34^{b}	37^{b}
RF 95 D21	$5,7^{c}$	47	50^{c}	41	47^{b}

Freshness decreased significantly from the first day of storage to the seventh and from seventh day to day 21. Then the freshness score was around 5,5 which has been used as the limits for shelf life at IFL (Figure 15). Both RF and CON heated samples were significantly firmer (lower score for softness) at day 21 compared with fresh cooked and reheated samples on the seventh storage day. RF samples were softer and more tender than other samples on day 7. Scores fore flakes were lower when compared to heat processing at 75°C. Here it was below 56 compared to 47-63 in experiment 3 were samples were heated at 75°C. More variation were within storage time in evaluated texture attributes in samples heated at 95°C than heated at 75°C.

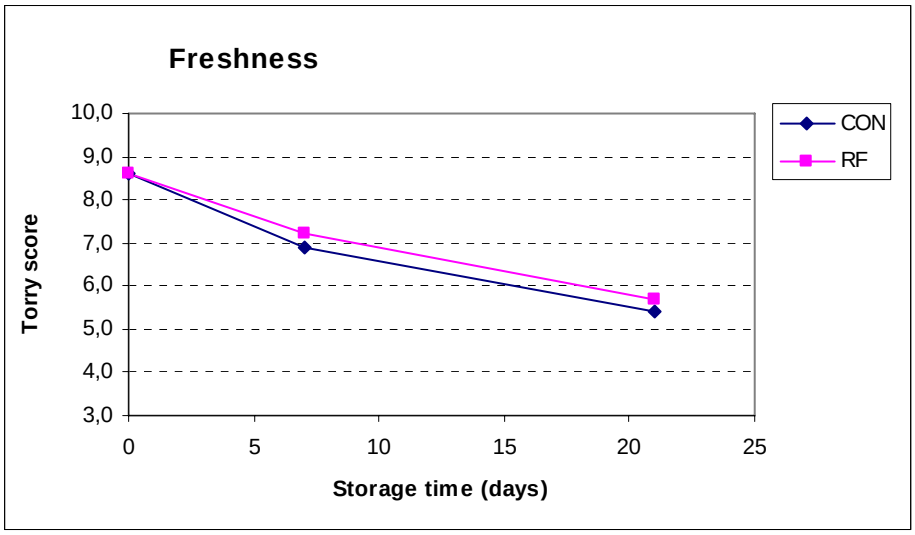

Figure 15. Average Torry scores of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL. Storage at 3°C

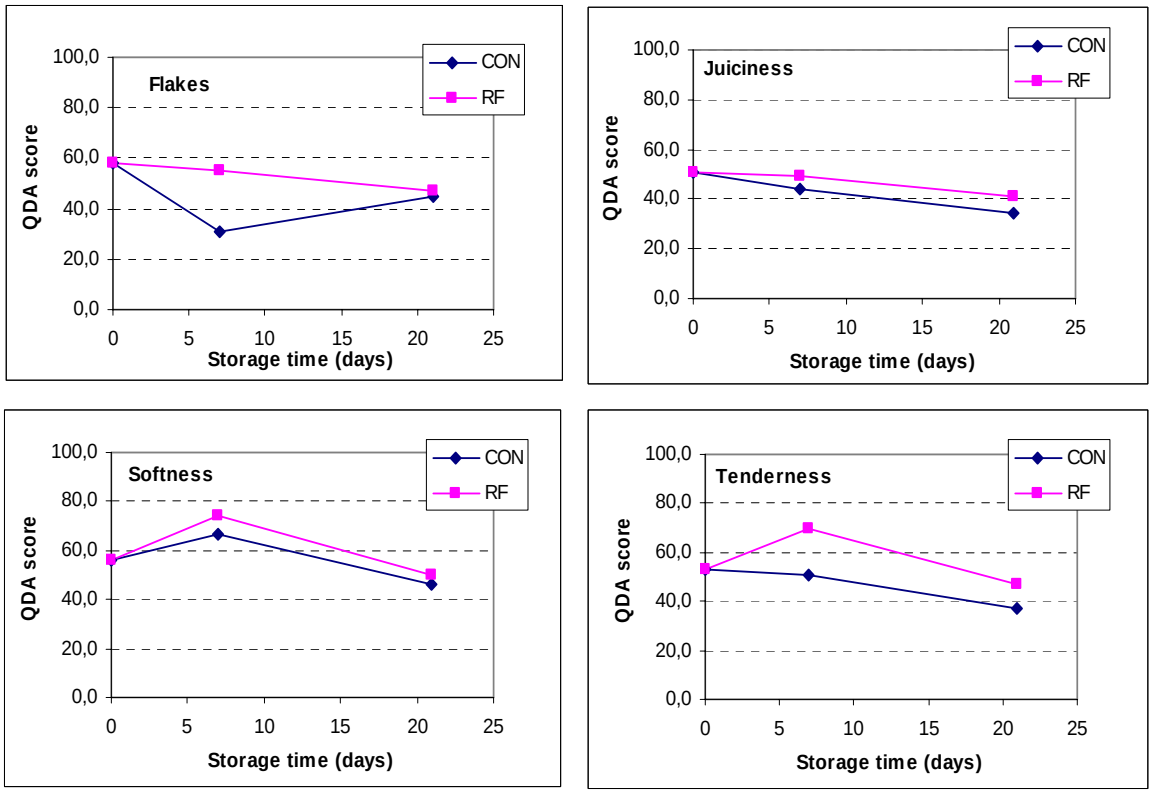

Figure 16. Average QDA (texture) scores of cooked/re-heated cod; RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod. On day 0, fresh cod was analysed after heating at IFL Storage at 3°C.

Cook-out and water holding capacity (WHC)

Cook-out in cod samples was not significantly affected by type of heat treatment but lower after 22 days of storage compared to 7 days. However, in previous trial (experiment 3) with storage of chilled samples it was observed that cook-out was higher in CON than RF-heated samples. Higher temperature during heat treatment in both categories may have resulted in less difference but the condition of the raw material may also be a factor worth considering. WHC was neither affected by heat treatment or time which confirmed results in experiment 3 (Figure 17).

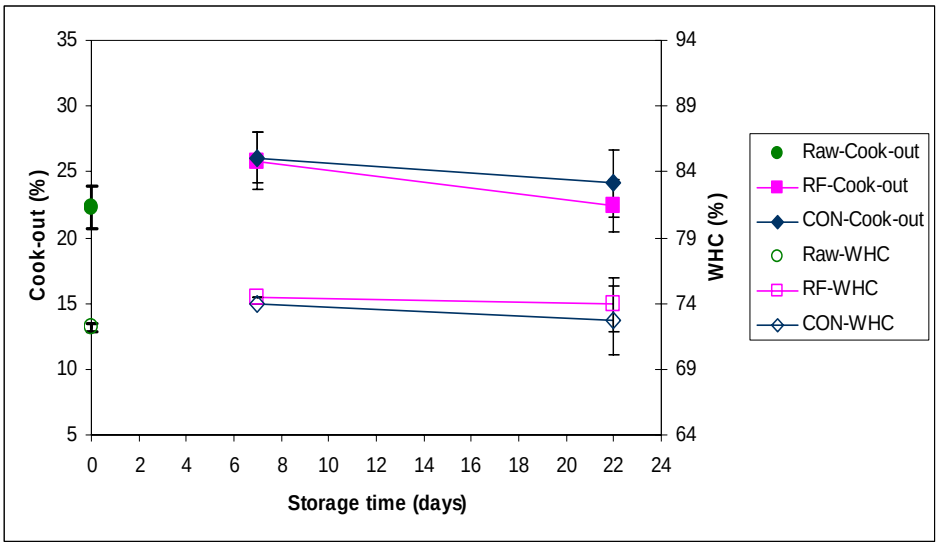

Figure 17. Cook out and WHC (%) of precooked samples (n=3) which were reheated after 7-22 days at 2°C; (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh cod was analysed after heating at IFL

Water content and pH

Effects of heat treatment on water content were not significant but it was higher after 22 days then after 7 days of chilled storage as could be expected with regard to results for cook-out. On the contrary, significant difference was found between CON and RF in trial 3, again the explanation may have been that higher temperatures during heat treatment have minimized differences between heat treatments. The pH was higher in CON-heated samples and increased from day 7 to 22 (Figure 18). The opposite was observed in experiment 3.

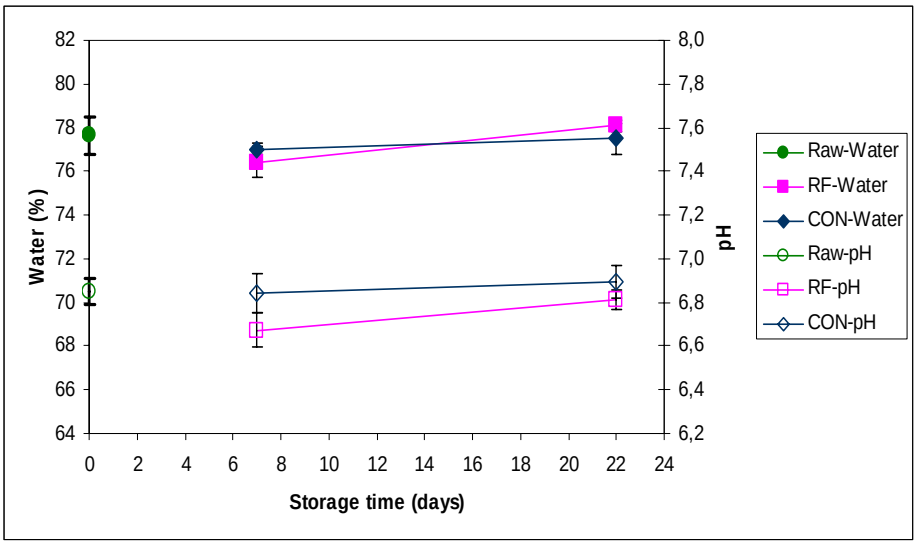

Figure 18. Water and pH of precooked samples (n=3) which were reheated after 7-22 days at 2°C; (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh cod was analysed after heating at IFL

Formaldehyde (FA) and thiobarbituric reactive substances (TBARS)

The formaldehyde content was 208.7 ± 115 µg/g in fresh single cooked samples on day 0 but 15.3 and 13.2 µg/g on day 7 in RF and CON reheated samples respectively. The value obtained for the fresh fish was low but the variation between individuals was high. The limit value of formaldehyde for human consumption has been reported to be 75 mg/kg in raw fish. Values of less than 10 mg/kg indicate that frozen products are of good quality (Rehbein 1987).

The TBARS was 1.1 ± 0.3 µmol/kg in fresh cod but increased with storage time in both RF and CON heated samples. After 7 days of chilled storage the values were 3.8 and 3.6 µmol/kg but 5.1 and 8.1 µmol/kg after 22 days for RF and CON heated samples, respectively (Figure 19). The results were different from the ones obtained in trial 3, where the TBARS of both groups was higher (10.9 and 11.5 µmol/kg) and more similar than in this trial. Values observed in our trial remained well below the limit for TBARS in raw cod of 19 µmol/kg (Connell, 1975).

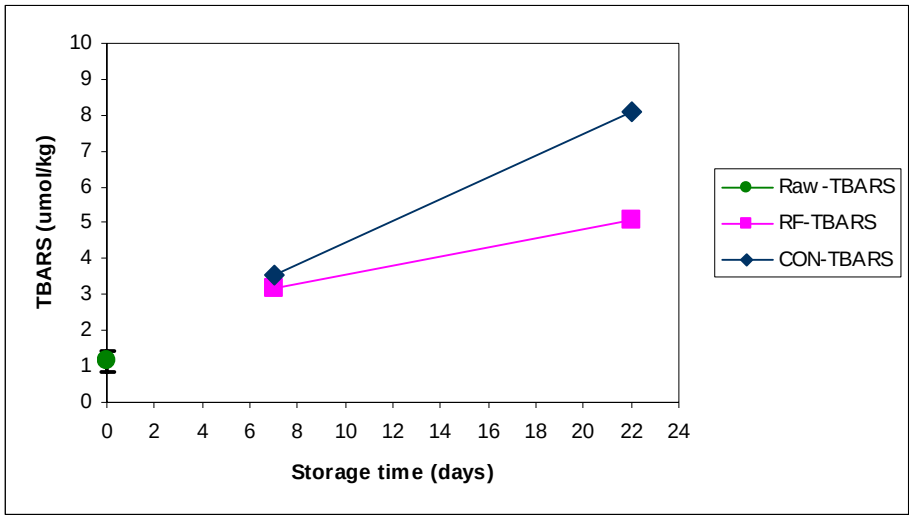

Figure 19. TBARS of precooked samples (pooled samples (n=3)) which were reheated after 7-22 days at 2°C; (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh cod was analysed after heating at IFL.

EXPERIMENT 6; SHELF LIFE OF CONVENTIONALLY (CON) AND RADIO-FREQUENCY (RF) HEATED AT 95°C SALMON STORED AT 3°C

Microbial counts

Total viable counts are shown in table 8. The TVC of the raw material was much higher than of the raw material used in experiment 4. However the number was within the limits defined for good quality in this project (Palsson et al 2005). The TVC measured in the pre-heated products was very low, indicating that the heat treatments were sufficient.

Table 8. Microbial counts (Total plate counts at 22°C) cfu/g.

Days after precooking	Sample	TVC
	Raw material	100000
day 6	RF	est<10
day 6	CON	est<10
day 20	RF	est<10
day 20	CON	est<10
day 28	RF	est<10
day 28	CON	est.10

Sensory evaluation of re-heated and cooked fillets

Samples were evaluated after 6, 13, 20 and 28 days at 3°C storage with sensory evaluation and raw material at same day as the heating process as day 0 in the shelf life experiment. The results are shown in Table 9 and Figure 20.

Table 9. Average sensory scores of salmon samples as evaluated by the sensory panel. Different superscripted letters indicate difference (p<0.05). CON; conventionally heated, RF; radio-frequency heated, RM; raw material heated at IFL, d=days in storage at 3°C, o = odour, f = flavour.

Sample	characteristic (o)	seaweed-sea (o)	liver-oil (o)	earthy (o)	sour (o)	rancid (o)	flakes	softness	juiciness	tenderness	characteristic (f)	metal (f)	oil (f)	sweet (f)	earthy (f)	sour (f)	rancid (f)
RM	54^{a}	38^{a}	32^{a}	35^{a}	8	4^{a}	60^{a}	58	59^{a}	62	60	45^{a}	44^{a}	30	28	10	8^{a}
CON95 d7	48	25	30	31	7	7^{a}	39^{bc}	59	50	55	50	38	37	25	38	9	15^{a}
RF95 d6	57^{a}	30^{ac}	27	16^{b}	2	5^{a}	51^{ac}	57	55	59	53	27	35	20	22	3	10^{a}
CON95 D13	40	19^{bc}	30	27	7	4^{a}	33^{bc}	61	46	52	45	25	35	26	30	8	11^{a}
RF95 D13	50	23	28	20	5	6^{a}	49^{ac}	59	53	61	52	29	34	29	27	5	6^{a}
CON95 D20	37	20^{bc}	20	33	9	8^{a}	39^{bc}	57	38^{b}	49	41	26	32	27	34	8	13^{a}
RF95 D20	42	23^{bc}	17	32	9	12	50^{ac}	54	43	55	46	26	36	25	43	13	21
CON95 D28	41	16^{bc}	20	38^{a}	8	23^{b}	45	60	49	53	41	21	32	21	41	13	34^{b}
RF95 D28	33^{b}	10^{b}	16^{b}	31	11	13^{a}	29^{b}	68	57^{a}	55	40	23^{b}	23^{b}	19	42	13	23

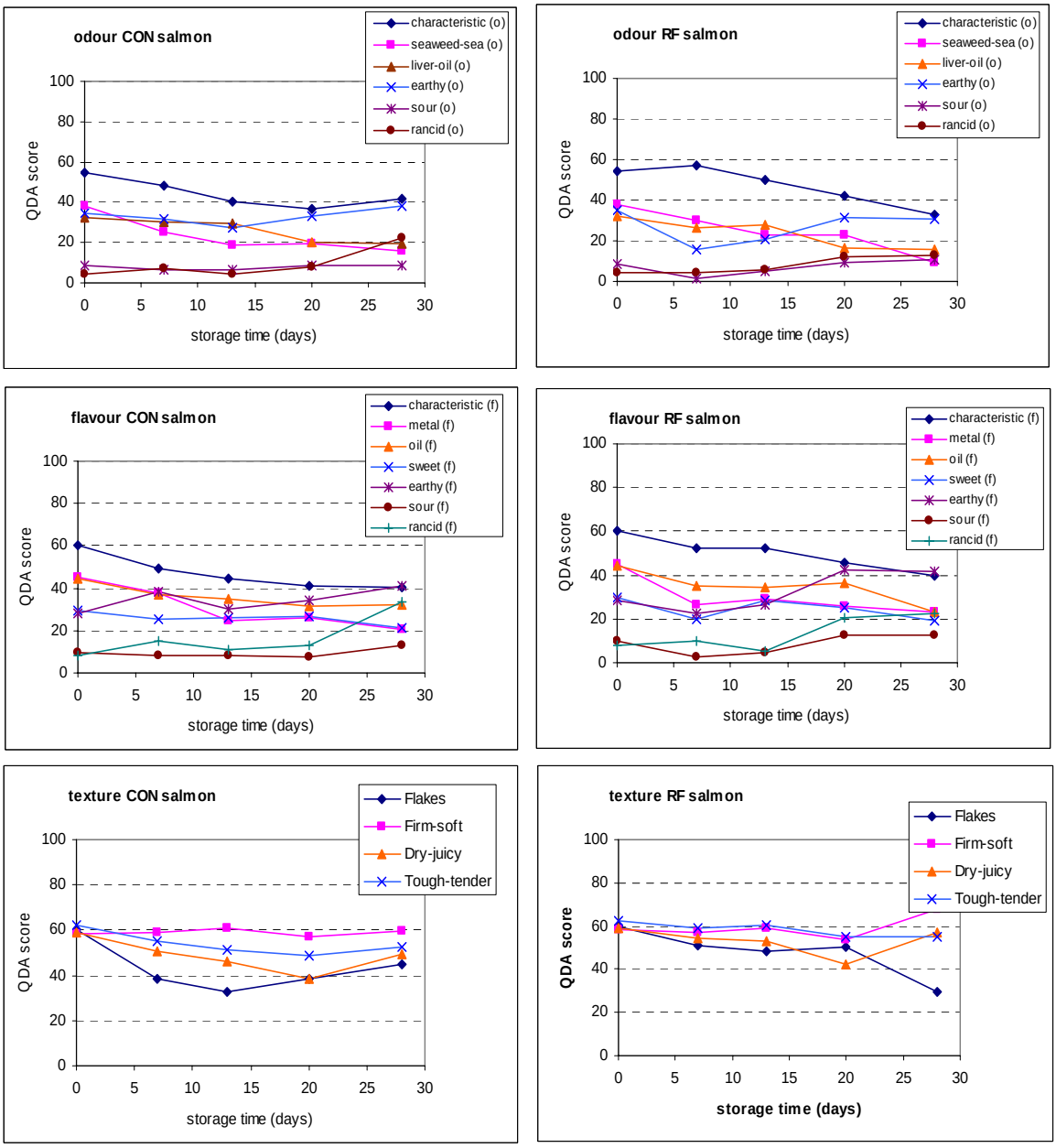

Figure 20. Average QDA (texture) scores of cooked/re-heated salmon; RF (Radio-Frequency heated at 95°C)-salmon and CON (Conventionally heated at 95°C)-salmon. On day 0, fresh salmon was evaluated after heating at IFL. Storage at 3°C.

Minor changes in sensory quality were observed during the first two weeks of chilled storage. Seaweed-sea odour had reduced significantly in CON heated samples after thirteen days. After twenty days seaweed-sea odour had reduced in both groups and rancid flavour increased in the RF samples. At storage day 28, CON samples were more rancid compared to RF samples. Texture did not change much during the storage time however it was significantly different in flakes with highest score in raw material and lowest in RF sample at day 28 (Table 9).

Compared to the shelf life experiment after 75°C intensity of rancid flavour was higher in samples heated at 75°C than samples heated at 95°C on day 20. On day 28 both sample groups had intensity score of rancid flavour higher than 20. Therefore it could not be stated that higher temperature in precooking prolongs the shelf life.

Instrumental texture measurements

Measured average values for hardness, cohesiveness and resilience are shown in Figure 21, 22 and 23 respectively. The RF heated samples had significantly more hardness than the CON heated samples at the sampling points of 20 and 28 days. This was not seen in the hardness values at day 20 when salmon was cooked at 75°C, but there the hardness for RF and CON is almost the same. This could possibly indicate that the 95°C RF heated samples become firmer with storage than the 75°C heated samples. The cohesiveness was significantly higher for the RF heated samples than the CON heated samples at the sampling point of 20 days. At the sixth day of storage the CON heated samples were significantly more resilient than the RF heated samples. However in sampling day 28 are the RF heated samples significantly more resilient than the CON heated samples. As mentioned before there was a lot of difficulty problems with the cooked salmon samples. It was difficult to cut them into sample sizes for the instrument and when compressed the samples tended to slide apart. The sampling preparation and measuring process was still more difficult for the 95 °C cooked salmon than the salmon that was cooked at 75 C. The fish structure keeps better when heated at lower temperature.

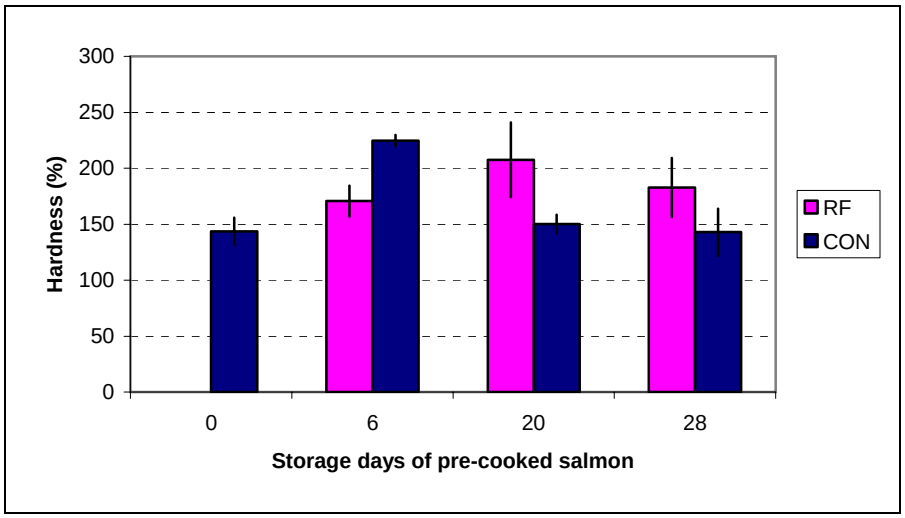

Figure 21. Hardness of cooked/re-heated salmon; RF (Radio-Frequency heated at 95°C)-salmon and CON (Conventionally heated at 95°C)-salmon. Calculated values with standard deviation from the time–force curve of TPA measurement.

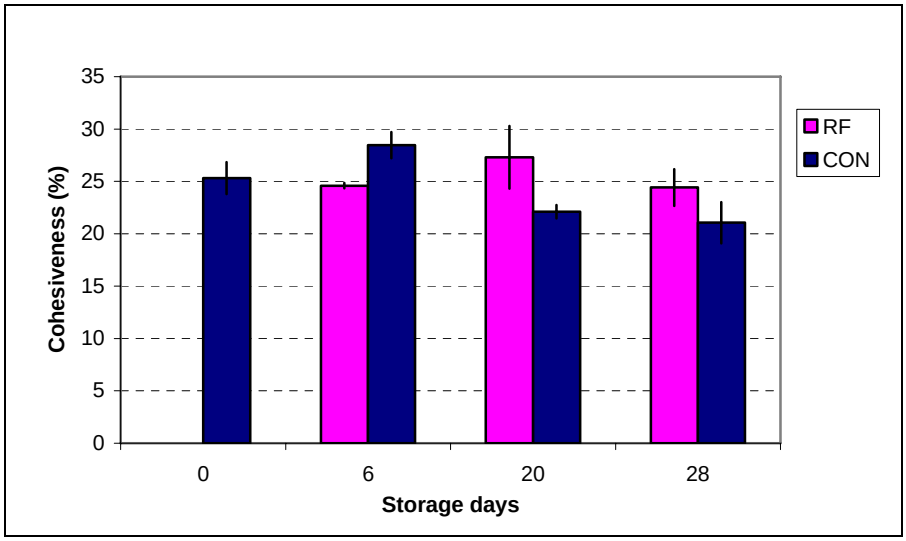

Figure 22. Cohesiveness of cooked/re-heated salmon; RF (Radio-Frequency heated at 95°C)-salmon and CON (Conventionally heated at 95°C)-salmon.. Calculated values with standard deviation from the time–force curve of TPA measurement. On day 0, fresh salmon was analysed after heating at IFL

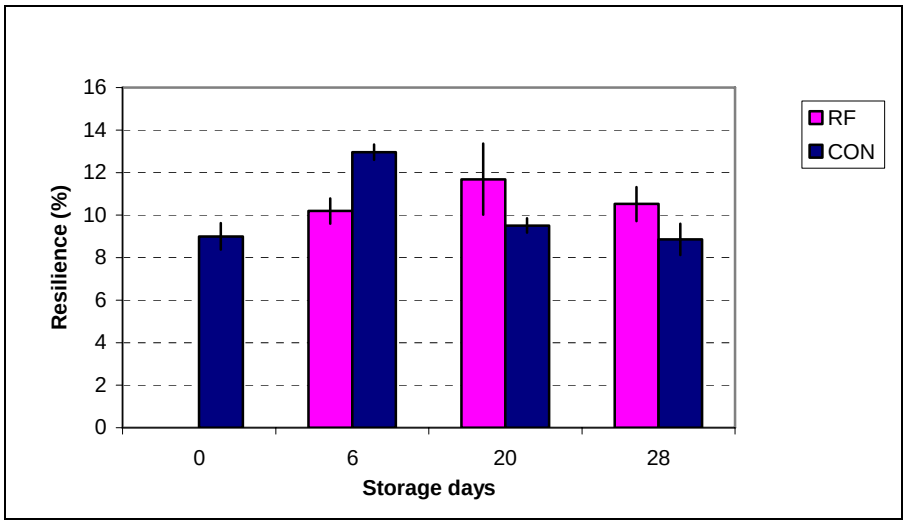

Figure 23. Resilience of cooked/re-heated salmon; RF (Radio-Frequency heated at 95°C)-salmon and CON (Conventionally heated at 95°C)-salmon. Calculated resilience (%) values with standard deviation from the time–force curve of TPA measurement. On day 0, fresh salmon was analysed after heating at IFL

Cook-out and water holding capacity (WHC)

Cook-out (%) was lower in the fresh salmon (6.1 ± 0.2%) than in CON (11.1-12.5%) and RF-heated samples (8.0-14.0%). It tended to decrease with storage time, more in RF-heated samples than in CON-heated samples (Figure 24). On the latest sampling day, the difference between RF and CON heated samples was significant.

WHC (%) was lower in the CON (59.3-66.1%) than in RF-heated (65.8-71.4%) samples. It tended to decrease with time except that samples on day 20 had higher WHC than on other sampling days.

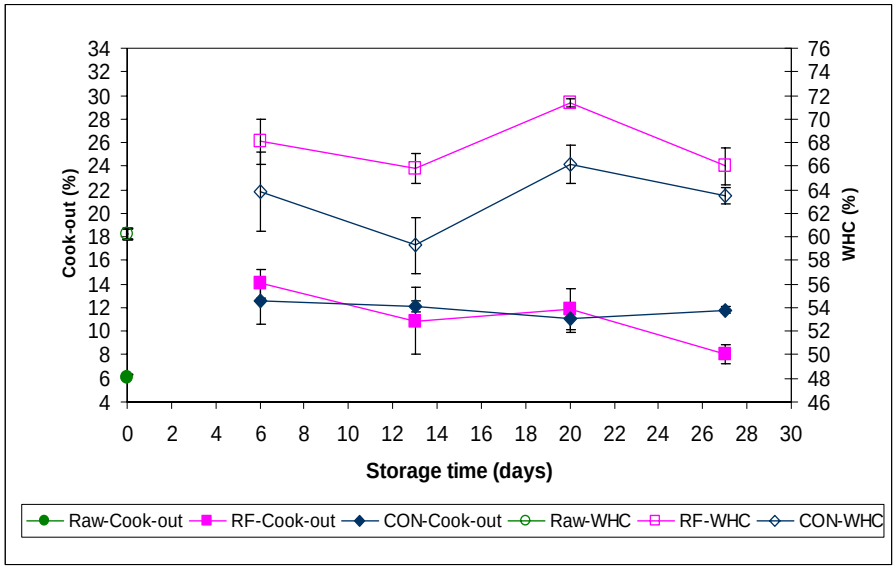

Figure 24. Cook out and WHC (%) of precooked samples (n=3) which were reheated after 6-27 days at 3°C; (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh salmon was analysed after heating at IFL.

Water and pH

The water content was not affected by heat treatment but was lower on day 13 than on other sampling days (Figure 25). On the contrary, water content was lower in CON samples in experiment 3.

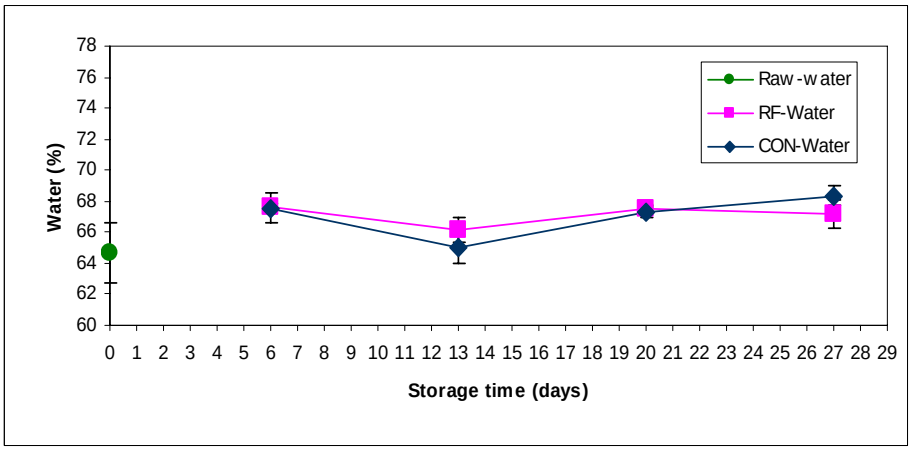

Figure 25. Water content (%) of precooked samples (n=3) which were reheated after 6-27 days at 3°C (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh salmon was analysed after heating at IFL.

The pH in fresh heated samples was 6.28 ± 0.03. It was not affected by heat treatment or time, although pH in samples on day 13 was slightly higher than on other sampling days (Table 1).

Table 10. pH of precooked samples (n=3) which were reheated after 6-27 days at 3°C (RF =Radio-Frequency heated and CON = Conventionally heated). On day0, fresh salmon was analysed after heating at IFL.

	6			13			20			27		
RF-pH	6.27	±	0.02	6.33	±	0.02	6.30	±	0.03	6.27	±	0.04
CON-pH	6.30	±	0.03	6.34	±	0.03	6.30	±	0.01	6.30	±	0.01

Peroxide value (PV) and thiobarbituric reactive substances (TBARS)

The PV in fresh heated salmon was 5.6 meq/kg. It was lower in CON on day 6 and 13, 3.1 meq/kg and 2.5 meq/kg, respectively but increased to 6.6 and 7.0 meq/kg on day 20 and 27 respectively. Peroxide values of RF-heated samples were higher on day 6 (7.4 meq/kg) and 13 (4.4 meq/kg) but slightly lower on day 20 and 27 (5.8 meq/kg for both days). The PV tended to increased with time for CON-heated samples but the opposite was observed in experiment 3.

The TBARS of the fresh salmon after cooking was 17.7 μmol/kg which indicated that oxidation had started and rancidity could be expected. After 6 days of chilled storage, the TBARS of the CON-heated samples (7.9 μmol/kg) was lower than of RF-heated salmon (16.4 μmol/kg). It maintained higher in RF heated samples although only slight differences were observed on the final day, 12.4 vs. 13.3 μmol/kg in RF heated samples. The highest values were observed after 20 days of storage but after 27 days of storage the values obtained were similar or slightly lower than on day 13 (Figure 26).

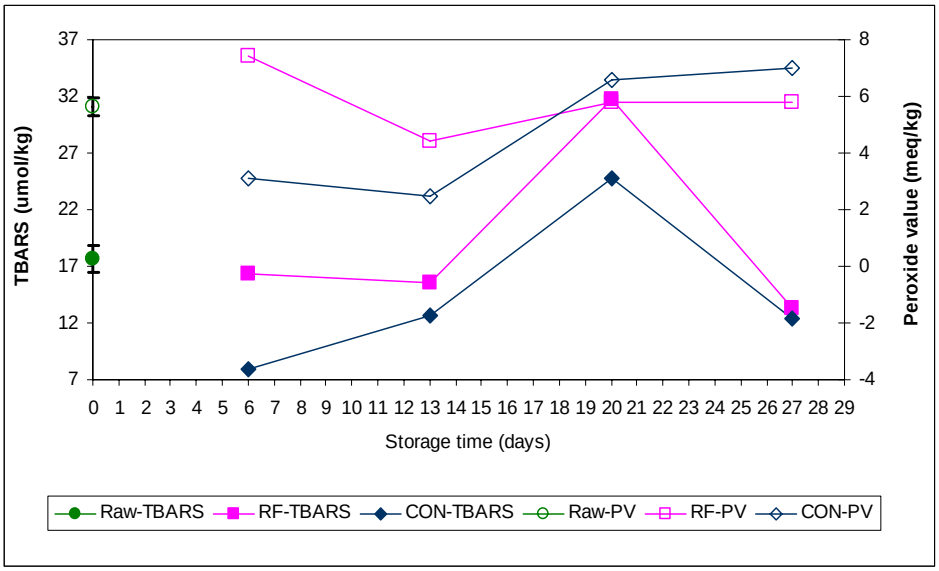

Figure 26. TBARS and PV of precooked samples (n=3, pooled on day 6-27) which were reheated after 6-27 days at 3°C (RF =Radio-Frequency heated and CON = Conventionally heated). On day 0, fresh salmon was analysed after heating at IFL.

Comparison of measurements

Results from all methods were averaged over sample groups and compared in a PCA plot for cod (Figure 27) and salmon (Figure 28) to compare methods and effects of temperature during heat treatments.

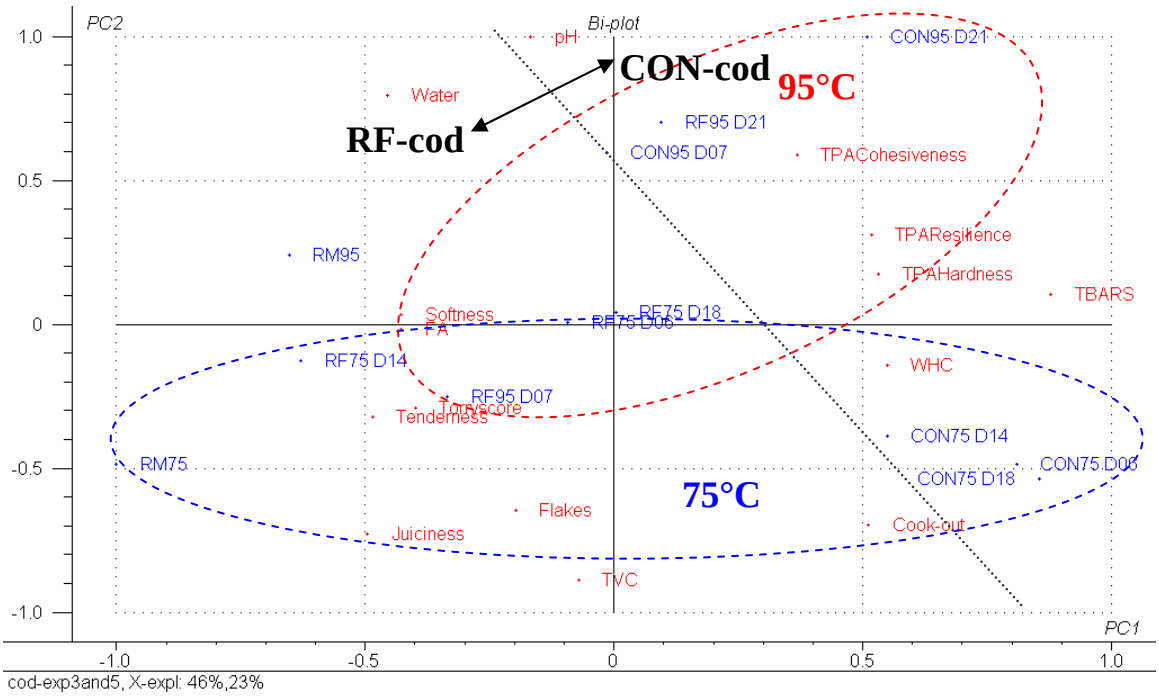

Figure 27. Scores and loadings (Bi-plot) of all measured parameters and sample groups, PC1 vs. PC2.; raw material, RF (Radio-Frequency heated)-cod and CON (Conventionally heated)-cod after chilled storage at 3°C

The first PC axis in the bi-plot (Figure 27) mostly appears to explain differences between cod samples due to storage time, freshness (Torry freshness score), to the left and TBARS to the right. Variation with regard to texture is also included in the difference between samples along the first PC. Textural parameters account for most of the variation between the samples along PC2.

Cod heated at 75°C appears to be different from cod heated at 95°C as the samples are separated in the bi-plot along the PC2 axis. The cod heated at 75°C appeared to be more juicy, tender and flaky compared to the 95°C heated cod.

Figure 27 also shows that there appears to be some differences between the RF and CON heated samples. Most RF-heated samples, along with the cooked raw material for both 75 and 95°C experiments, are located in the left side of the plot and the CON-heated samples to the right. According to this it appears that the re-heated RF-heated samples are more alike cooked raw material used, with more juicy, flaky, tender and soft texture, while the CON-heated samples have somewhat more WHC and higher values of TBARS.

The measurements gave various information about the samples. As might have been expected, samples that had more juicy texture were opposite to samples with high cook-out on the PC1 axis. Samples with high freshness scores were also more juicy, flaky, soft and tender and opposite to instrumental texture parameters on both axes, which might be expected, as hardness and resilience might be regarded as the opposite of soft and tender.

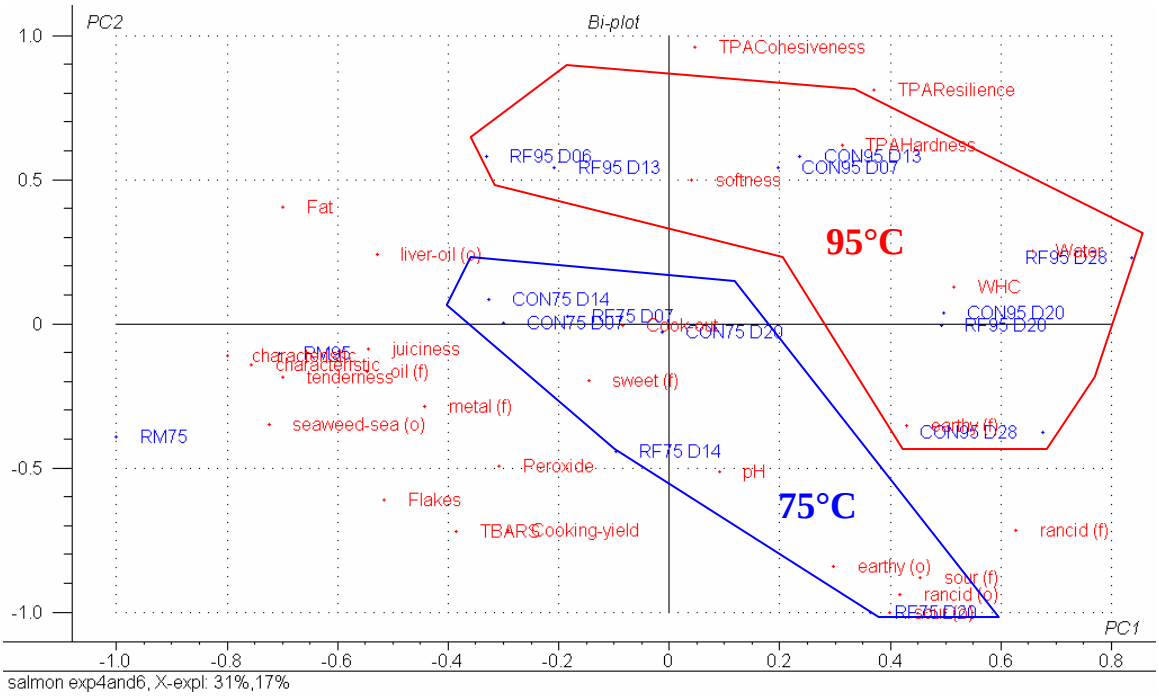

Figure 28. Scores and loadings (Bi-plot) of all measured parameters and sample groups, PC1 vs. PC2.; raw material, RF (Radio-Frequency heated)-salmon and CON (Conventionally heated)-salmon after chilled storage at 3°C

Similar trend was observed for the salmon samples in Figure 28 as for the cod samples in Figure 27, in that the first PC axis appears to show differences between samples with regard to freshness, but the second PC axis is more related to texture. Also, the salmon

pre-cooked at 75°C is different from the salmon pre-cooked at 95°C. The 75°C salmon is located in the lower side along with the cooked raw material used for both experiments, but the 95°C salmon is in the higher side of the plot. The 75°C salmon appeared to be more flaky and tender, while the 95°C salmon was measured with more resilience and more hard texture. The 95°C group appeared to be more to the right, with more samples described with sensory spoilage descriptors such as sour odour and flavour.

4. CONCLUSIONS

The quality and shelf life of cod and salmon, pre-heated with two different heating processes at two different temperature, and stored at 3°C was studied.

In freshness evaluation of reheated cod samples there was no significant different by heat treatments (RF vs CON) neither at 75°C nor at 95 °C samples. Off flavours was detected after fourteen days of cod samples pre-heated at 75°C.
After twenty days of storage, intensity of rancid flavour in pre-heated salmon was above acceptable limit. Therefore shelf life could not be estimated longer than that twenty days of storage after both 75°C or 95°C pre-heating treatments.

Texture of both cod and salmon samples was not affected by storage time. Higher temperature (95°C) during pre-heating influenced the texture negatively and it became more difficult to evaluate and measure the texture. Fish pre-heated at 75°C was more juicy, tender and flaky and retained its form better compared to fish pre-heated at 95°C.

Sensory evaluation of salmon using the Quantitative Descriptive Analysis gave valuable information about odour, flavour and texture of the salmon products with storage time and if there were differences between products. For cod, only the Torry freshness score sheet was used and some additional descriptors for texture. Even though the Torry freshness score sheet has been used frequently for fresh and thawed cod, it only gives information about freshness in general, which it also did here. However, some more detailed information about odour and flavour would have been useful and could have given more valuable information about the detected off-odour and flavour of the cod.

Overall, the results indicate that radio-frequency heated cod and salmon are of comparable shelf life and/or better quality compared to conventionally autoclave heated cod and salmon both during frozen and chilled storage. However, it must be mentioned that the shelf life studies with the radio-frequency heated fish were on a product generated with not fully developed equipment for radio-frequency heated fish. That is to say, the production process and packaging process had not been fully developed and the

quality and shelf life studies can only give quality indications about the final product, produced during a real product production.

It is clear that shelf life is not only influenced by the production technique used to heat the product. Other factors are of high importance, such as the raw material used, the fish species which may react differently to the process, the production process, the packing material and storage temperature.

5. ACKNOWLEDGEMENTS

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APPENDIX A: SHELF LIFE STUDY ON CHILLED COD IN GREEN ONION SAUCE

Introduction

In October/November 2004, pre-cooked cod (conventionally and radio-frequency heated) was evaluated by the Icelandic Fisheries Laboratories (IFL) sensory panel. The results showed that the cod had reached the limits for acceptability (end of shelf life) after 13 days of chilled storage at 3°C.

This was very different from the given shelf life of pre-cooked cod products produced and sold by Fjordkökken. According to the sensory panel at Fjordkökken, chilled pre-cooked cod products had a shelf life of at least 28 days.

The explanation for those different results may be due to that at Fjordkökken, the whole product was evaluated, but the sensory panel at IFL evaluated the pre-cooked cod samples without any additives or trimmings. In addition, the raw material may have been different.

Because of those different results regarding the determination of maximum shelf life, it was decided to carry out and additional experiment in the RF-project. The aim was to make the two panels evaluate the same product (cod in green onion sauce), produced at Fjordkökken and compare the evaluation of freshness quality.

Materials and Methods

Cod in green onion sauce

Cod (deskinned, pieces of fillets without bones) in green onion sauce, produced at Fjordkökken. The ingredients in the sauce were green onion, milk, water, salt and flour.

Cod fillets were thawed in a chilled chamber for 2 days. Around 140 g of thawed cod pieces were packed together with green onion sauce (135g) and heated in plastic bags in boiling water for 5 min at 95°C. Each pack weighed around 275g.

After production, samples for evaluation by the sensory panel at Fjordkökken were stored at 3-4°C at Fjordkökken, and samples for evaluation by the sensory panel at IFL were transported (packed in polystyrene boxes with ice mats) by flight to IFL and stored at 3-4°C.

Samples were evaluated 4 times during 4 weeks (8, 12, 19 and 26 days after heat treatment at Fjordkökken).

Sensory evaluation

The product was evaluated on same days by the two panels.

The method used by the IFL panel was a 5 point grading scale from 5 (very good) to 1 (unacceptable) for freshness and intensity scale for spoilage odour and flavour from 0 (none) to 4 (very much spoilage odour/flavour) (Table 1 and 2 show the scales used by the IFL sensory panel). Nine to ten panellists participated in the sensory evaluated each time and samples were evaluated in duplicates (coded with random digit numbers that did not indicate the storage time or any information about the product). The panel was not trained specifically for the evaluation of this product, but were all very experienced in sensory evaluation of cod, and were familiar and trained in using the scales used in the test.

The method used by the Fjordkökken panel (2 expert panellists) was also a 5 point grading scale from 5 (right quality), 4 (satisfied quality), 3 (should be changed to better), 2 and 1 (poor quality).

The parameters evaluated at Fjordkökken were as follows:

- Appearance; light, thick with a green appearance from the onion, no skin and bone and should be 1 whole piece of fish, will accept 2 pieces
- Texture; very little chewing resistance, sauce should be thick and smooth
- Flavour: Fish should have a clean good fish taste, sauce should taste of fish bouillon, green onion and salt

The sensory evaluation was carried out the usual way at Fjordkökken; two panellists participated in the sensory evaluation, with expert knowledge of the product.

Table 1. Freshness score sheet used by the IFL panel to evaluate the freshness of cod in green onion sauce.

Description		Score
Very good	Watery, metallic, sweet, meaty, characteristic for the species	5
Good	Sweet, less characteristic	4
Acceptable	Neutral	3
Questionable	A little sour, hint of off-flavour	2
Unacceptable	Sour, bitter, TMA, off-flavour	1

Table 2. Intensity scale used by the IFL panel to evaluate spoilage characteristics of cod in green onion sauce.

Score	Description
0	None detectable
1/2	
1	Hint
2	Little
3	Obvious
4	Very much

Data analysis

The samples were compared in NCSS 2000 (NCSS, Utah, US), with one way ANOVA, multiple comparison was done using Duncans´ test. Significance level was set at 95%.

Results and discussion

Figure 1 shows the results from the IFL sensory panel.

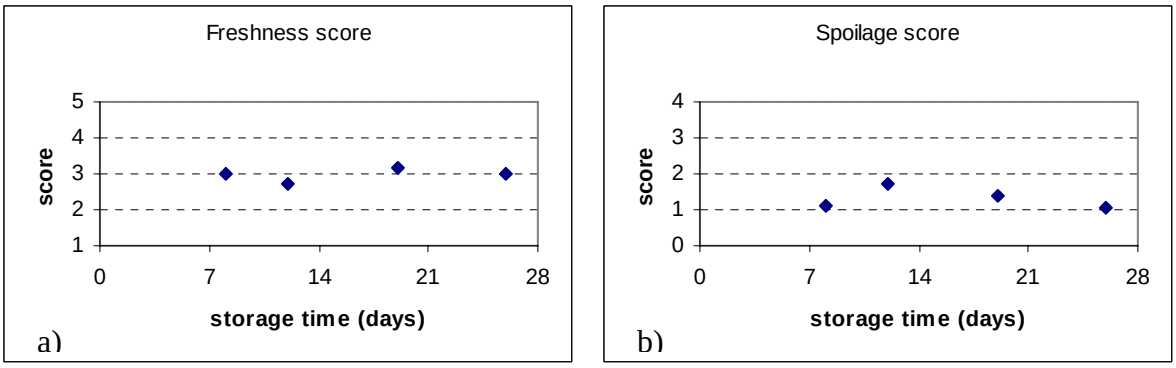

Figure 1. Average freshness (a) and spoilage (b) scores for cod in green onion sauce, based on the sensory evaluation of the IFL sensory panel.

No significant difference was found between the samples with regard to freshness or spoilage odour/flavour. The freshness of all samples was around 3 (neutral) during the storage time, and spoilage odour/flavour was scored around 1 to 2 through the storage time (hint to little spoilage odour/flavour). Around half of the panellists commented on frozen storage odour/flavour and tough texture in all sessions, which may have influenced the freshness and spoilage scores. In the last sessions the panellists also commented on discolouration, green colour of the cod. Therefore, the products might be considered to be at the limits of acceptable quality after 26 days of chilled storage.

The results from the Fjordkökken sensory panel showed that the quality was very good the first 3 weeks of storage (freshness score = 5) for all evaluated attributes, which was not in accordance with the IFL panel. However, the Fjordkökken panel results during the 4^{th} week were not different from the results of the IFL panel, scoring appearance = 3, texture = 4 and taste = 4, also detecting green discolouration of cod.

Conclusions

The results from the IFL sensory panel indicated that the raw material used in the product "cod in green onion sauce" was not of high freshness quality. The average freshness score indicated that the quality was only "acceptable" from week 1 and until week 4, with remarks on tough texture and frozen storage odours and flavours. The description of the raw material used in the product showed the raw material had been frozen before product production. Therefore are the result from the IFL sensory panels evaluation of the product

not surprising with regard to comments on frozen storage odour/flavour and tough texture. However the incongruity between the panels may be caused by the fact that the freshness scales used to evaluate the product were quite different with regard to descriptions, including hedonic descriptions in the scale used by the Fjordkökken sensory panel. Also, it could be that the quality of the raw material used may be the standard raw material quality used by Fjordkökken and therefore evaluated as "right quality". Even though the cod in the product had frozen storage odours and flavours and tough texture, it is still acceptable quality for this particular product as it is not exactly spoilage.

Based on the results, 26 days might be considered as maximum storage time for the chilled product "cod in green onion sauce", mainly due to discolouration of the cod.