Textural Properties of Raw Salmon Fillets as Related to Sampling Method

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ABSTRACT

Textural properties of raw Atlantic salmon (*Salmo salar*) fillets from different origin were studied on different locations of the fillets. Three instrumental methods were applied for evaluation of textural properties. Two methods were based on puncture tests, using flat-ended cylinder or spherical probes measuring the hardness of the fillet. The third method was based on cutting the fillet with a blade and measuring the shear force. Hardness and shear force increased from head to tail, and the location below the dorsal fin was found to be most reliable. The shear force method was found to be more sensitive than the puncture methods and best suited for practical application.

Key Words: Atlantic salmon, texture, sampling method, shear force, hardness.

INTRODUCTION

THE MAIN QUALITY PARAMETERS FOR FRESH SALMON ARE FAT, color, texture and freshness. Other parameters commonly cited are white stripes (connective tissue), bloodstains, marbling and melanin (Koteng, 1992; Sigurgisladottir et al., 1997). Texture of raw salmon fillets is commonly tested in the industry by the "finger method". A finger is pressed on the skin or the fillet and firmness is evaluated as a combination of the hardness when pressed on the fillet and mark or hole left in the fillet after pressing. This method depends to a large extent upon subjective evaluation of the person who is performing the measurements (Sigurgisladottir et al., 1997).

Texture of raw salmon fillets may be measured objectively by different methods using mechanical food testing equipment (Instron). The main techniques applied for fish may be classified into puncture, compression, shear, and tensile techniques. A variety of shearing and cutting devices are available such as Warner-Bratzler and Kramer Shear Compression Cells. The main disadvantage with the Kramer devices is that they require large samples and a nondestructive method would be more favorable. Double compression makes it possible to perform a the texture profile analysis (TPA) from a plot of force-time curves (Bourne, 1978). Other terms use to describe texture are firmness, stiffness and yield point (Borresen, 1986; Botta, 1994, Andersen, 1995). When using instrumental methods, such measurements are limited by the instrumental behavior of materials in terms of stress, strain and time effects. Many attempts have been made to correlate physical measurements with sensory evaluation of texture (Breene, 1975; Borderias et al., 1983; Karl and Schreiber, 1985; Ragnarsson, 1987; Botta, 1991; Durance and Collins, 1991; Johansen et al., 1991; Reid and Durance, 1992; Chamberlain et al., 1993).

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Reproducibility of texture measurements is affected by sampling technique because of the heterogeneity of the fillets (Borresen, 1986; Botta, 1991; Reid and Durance, 1992). Therefore, it is difficult to find a representative average sample and measurements of textural properties may depend on the location within the fillet. However, raw fish should be tested in the form of a fillet or a part of a fillet. The chemical composition of salmon can affect the perception of color and texture (Dunajski, 1979; Christiansen et al., 1995). Measurement of quality parameters gives variation along the salmon fillets from head to tail. Fat, pigments and collagen are distributed differently throughout salmon fillets. The variations of fat can range from 9.6 to 38% within a slice of a whole fish (Aursand et al.,1994). Texture of fish fillet is also related to the diameter of the muscle fibers. The strength is higher with smaller diameter and, therefore, higher numbers of fibers, than with larger diameter and lower numbers of fibers (Hatae et al., 1990). Thus sampling is an important factor in the evaluation of salmon fillet texture.

Sampling techniques differ widely in the salmon industry and samples may be collected from different areas of head to tail depending on country, research institute, or company. A standard is available in Norway on methods to determine color and fat content in salmon fillets for both sampling technique and methodology (Norwegian General Standardizing Body, 1994). However, no standard is presented for texture (Sigurgisladottir et al., 1997) and there is a need for standardization of a method and sampling technique for texture measurements. Textural properties of raw salmon fillets of different origins, such as comparison of farmed and ocean ranched salmon have not been published.

Our study was designed to compare objective methods and sampling techniques for evaluation of texture of fresh salmon fillets of different origins. Three methods were applied on different locations of salmon fillets. This study was a part of a co-operative development between several research institutes and the industry in Iceland, Norway, Danmark, and Faroe Island on methodologies and sampling techniques for measuring quality parameters of salmon fillets.

MATERIALS & METHODS

Sample preparation

Fish were bled immediately after collection from tanks and then iced. The fish samples were stored for 3 days after slaughtering on ice in sealed boxes in a refrigerator at 4°C. Samples was prepared at the Technological Institute of Iceland on the fourth day. The fish were filleted and the skin removed. Each fillet was cut into 7 parts (location 1–7, Fig. 1). All measurements were performed above the lateral line on the fillets.

Evaluation of sampling procedures and sample sizes

Samples of 50 salmon were used. A land based salmon farm in Iceland supplied fresh Atlantic salmon (4 kg). The study was split into two parts within 2 wk time, due to analysis limitation and 25 fish were used in each experiment. The salmon samples for both experiments were obtained from the same tank, with equal starvation time, and treated in the same way until measured.

Samples of different origin

Salmon (15 samples) of different origins were collected for each experiment. They were collected from sea-cages, land-based (tanks) farms and ocean-ranched fish from different locations (Norway, Iceland and Faroe Island). The fish samples were packed on ice in sealed boxes and delivered to the Technological Institute of Iceland by air within 3 days after slaughtering. Samples were prepared at the Technological Institute of Iceland as before. In this study, two attachments, sphere and a blade were tested on seven different locations on salmon fillets from head to tail (Fig. 1) to evaluate the textural properties.

Textural measurements

The TA.XT2 Texture Analyzer was used (Stable Micro System, Surrey, England). Three different attachments were applied, flat ended cylinder, spherical probe and a blade attachment.

Instruments

Results were based on application of the TA.XT2 texture analyzer with a load cell of 25 kg (Stable Micro System, England). This instrument provides a rigid framework for tension compression cycling and texture tests to generate true 3-dimensional product analysis of force, distance and time. Three different attachments was applied (Fig. 2: a, flat-ended cylinder; b, blade; and c, sphere) and various methods were tested for each attachment.

Flat-ended cylinder

Flat-ended cylinder of 25 mm diameter was selected to simulate the human finger. Constant penetration depth was applied on the fillets which were tested in the range of 4–6 mm. Penetration depth of 5 mm into the fillet was selected as the maximum distance which could be applied without breaking the muscle fibers and affecting the muscle structure by erupting it and leaving a mark on the fillet. Compression curves were compared at location 3 (below the back fin) and location 7 (tail), (Fig. 3) and the breaking point or yield point was at 6–8 mm
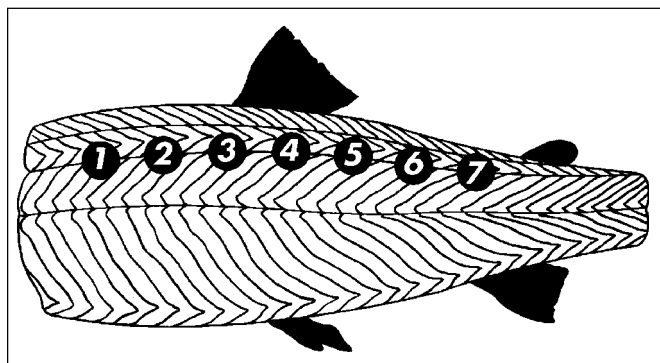
into the fillet. Double compression was applied to construct the texture profile analyses (TPA) parameters. The flat-ended cylinder approached the sample at the speed of 2 mm/sec and penetrated 5 mm into the fillet. Then the force was reduced and the fillet was allowed to rebound 15 sec with the cylinder just touching the surface. Then the cylinder was pressed on the fillet a second time and TPA was obtained by analyzing the force time curve (Bourne, 1978). The hardness was the height of the first peak.

Blade

The blade (knife edge, 60°) had a thickness of 3.0 mm and width of 70 mm which cut through the sample at a speed of 2.0 mm/sec. The shear force was measured as the maximum force required to shear/cut through the samples, i.e., the peak height. The blade approach was applied by pressing the blade through the muscle vertical to the muscle fibers. Thickness of salmon fillets varies from head to tail but is 2 cm thick above the lateral line. The samples were all cut into pieces of equal size, 2 cm in thickness and 4 cm in diameter. For precision and accuracy of results when using the cut approach all samples had to be equally thick.

Spherical probe

A sphere was selected as the second probe to simulate further the human finger method. It was applied on both fillets of natural thickness and samples made equally thick. The sphere affected the muscle less than the flat cylinder when pressed on the fillet. Therefore, it was possible to press 5-6 mm depending on the thickness of the fillet without breaking the muscle fibers and 5 mm distance was chosen. The locations closer to the tail part were more sensitive to breaking than the front part of the fillet. Double compression was applied as for the flat ended cylinder. The spherical probe was 25.4 mm in diameter. The same procedure was used as for the flat-ended cylinder.

Statistics

Data sets were compared by multiple comparison ANOVA using all pair wise comparison by Sigmastat 2.0 (Jandel Scientific Software, Ontario, Canada). Significance of difference was defined at $P<0.05$. Unscrambler analytical software was used to perform a 3-way (time period x method x fillet thickness) multiple ANOVA. The coefficient of variation (CV), calculated for each method (cylinder, sphere and knife), each location (1–7), and across both sampling periods was modeled with the aid of partial least squares (PLS) regression (Martens and Nas, 1987). The CV represented the sum of natural variations between fish and the variations related to specific methods and fillet locations (Nortvedt et al., 1996).

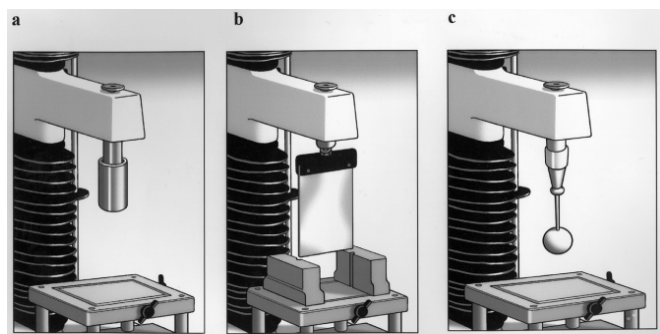**Fig. 1—Textural measurements were performed on locations (1-7) marked on the fillet.**

Fig. 2—Attachments applied for textural measurements of salmon fillets: (a)flat cylinder with diameter of 25 mm; (b) blade with thickness of 3.21 mm and width of 70 mm; and (c) sphere with diameter of 25.4 mm.
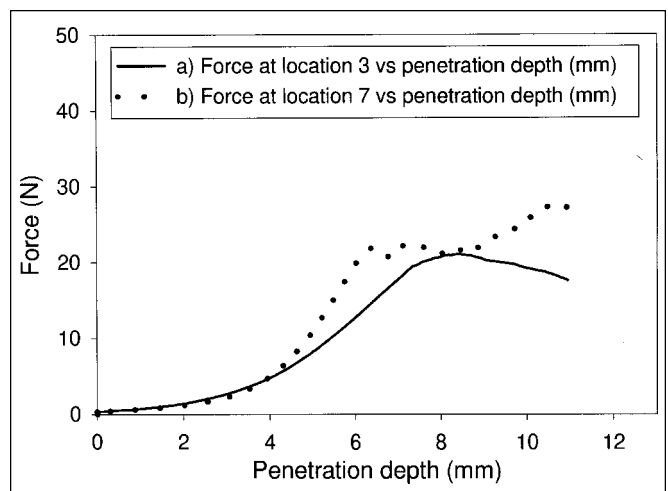

Fig. 3—Example of compression curves through fresh salmon fillets, where force is plotted against distance into the fillet from the surface. (a) Cylinder applied at location 3; (b) cylinder applied at location 7.

The following equation (1) was used for estimation of sample size:

$$n \geq 2 \cdot (CV/ES)^2 \cdot (t_{\alpha[v]} + t_{2 \cdot (1-P)[v]})^2 \qquad (1)$$

ES is the effect size, the smallest true difference between groups that is desired to detect, expressed as % of the mean, CV is the sample coefficient of variation, α is significance level (0.05), v is degrees of freedom of the sample standard deviation with a groups and n replications per group, P is desired probability (power of the test = 0.8) that a difference will be found to be significant (if it is as small as ES) and t is a value from a two-tailed t-table with v degrees of freedom and corresponding to probabilities of α and $2(1 - P)$.

RESULTS & DISCUSSION

Sampling

Shear force and hardness recorded from both the flat cylinder method and the sphere were not different (P<0.05) between the two samplings (Fig. 4, 5, 6, 7 and 8). Results from the two samplings

were, therefore, treated as one group of data (n=50). The thickness of salmon fillets (4 kg) varied from head to the tail. The difference in thickness was from 3 cm at the front part to 2 cm thick at the tail, (Table 1). Shear force was measured by cutting fillet samples with a blade and shear force measured using the blade increased from head to tail (Fig. 4). Shear force was higher at locations 6 and 7 than locations 1 to 4 and also higher at location 5 than at locations 2, 3 and 4 (Fig. 4). Shear force was the same at locations 2, 3 and 4, which is the area below the back fin (Fig. 1). Locations 2 and 3 could thus be used as duplicates for shear force measurements on the same fillet.

These results can also be seen on the score plot (Fig. 9) where all methods were used for all samples. Locations 3, 4 and 5 were clearly separated from the other locactions, indicating they were similar in shear force and hardness by the three methods (Fig. 9). Standard deviation was higher at the tail part or at location 5, 6 and 7 than at other locations. Generally higher standard deviation would be a disadvantage for a method but possibly it maybe more descriptive on textural properties, because it reflects the variations between samples. Although, the samples measured for shear force were of equal
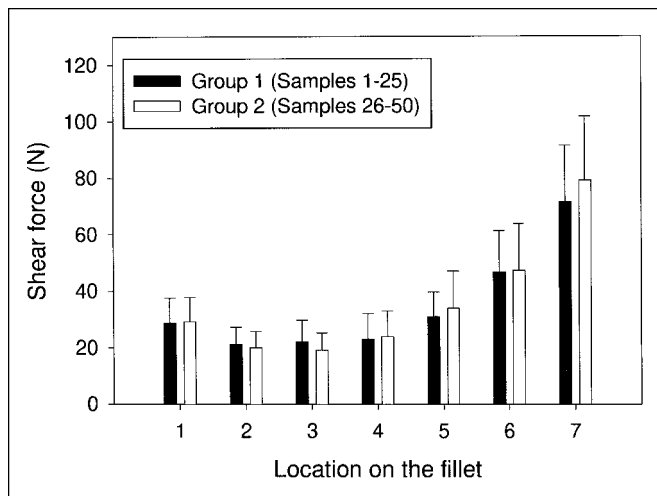

Fig. 4—Shear force of salmon fillets measured by cutting with blade at seven locations from the head to the tail. Data are mean and standard deviation of 25 fish from the first sampling (1-25) and the second sampling (26-50). Location 6 and 7 are significantly different from locations 1, 2, 3, 4 and 5.
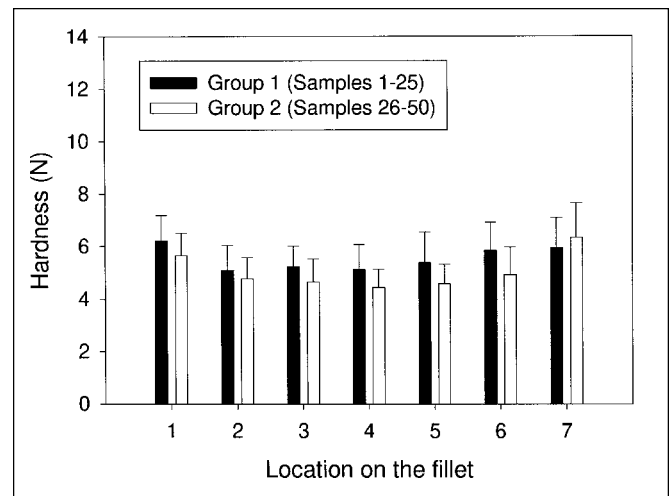

Fig. 6—Hardness of salmon fillets measured by using puncture with sphere at seven locations from the head to the tail. Samples were all equally thick. Data are mean and standard deviation of 25 fish from the first sampling (1-25) and the second sampling (26-50). Location 7 is significantly different from locations 2, 3, 4, 5 and 6.
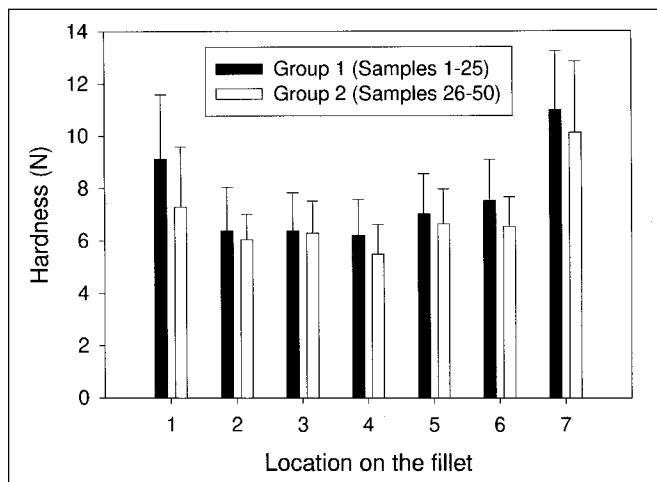

Fig. 5—Hardness of salmon fillets measured by using puncture with flat ended cylinder at seven locations from the head to the tail. Samples were all equally thick. Data are mean and standard deviation of 25 fish from the first sampling (1-25) and the second sampling (26-50). Location 7 is significantly different from locations 1, 2, 3, 4, 5 and 6.
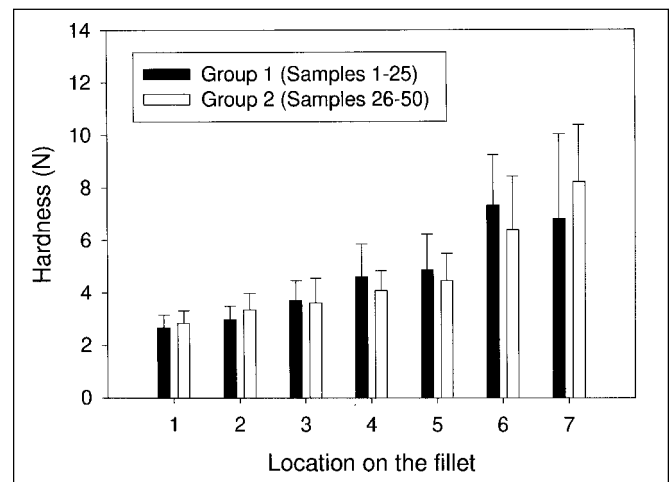

Fig. 7—Hardness of salmon fillets measured by using puncture with flat ended cylinder at seven locations from the head to the tail. Samples were naturally (not equally) thick. Data are mean and standard deviation of 25 fish from the first sampling (1-25) and the second sampling (26-50).Location 6 and 7 are significantly different from locations 1, 2, 3, 4 and 5.

thickness, the shear force increased from head to tail, with decreasing thickness of the original fillet. When the thickness of original samples was correlated to the results from shear force measurements, a correlation coefficient of 0.78 (R^2) (Table 2) was found. This, indicates the influence of chemical composition and structures of the muscle from head to tail on shear force (Fig. 4).

Hardness increased from the front part of the fillet to the tail using muscle samples made equally thick, both when flat cylinder and sphere were applied (Fig. 5 and 6). Hardness was higher at location 7 than at other locations, and location 6 was also higher than location 4. Hardness was, higher at location 1 than at other locations except for the tail part at location 7 (Fig. 5 and 6). The tail part of the salmon fillet was, therefore, found to be both more sensitive for breaking and higher in hardness than other locations (Fig. 3).

The thickness of the fillets varied from head to tail (Table 1) about 3 cm to 2 cm thick, but muscle samples from location 1, 2, 3, and 4 were not of different thickness.

Hardness of fillet samples of natural shape were not different between locations 2, 3 and 4 measured by cylinder or sphere (Fig. 7 and 8). More difference in hardness from head to tail was observed when fillets of natural thickness (Table 2) were used than when the fillet samples were made equal in thickness (Fig. 5, 6, 7 and 8). These results indicated that differences in thickness of the fillets from 2 cm to 3 cm affected the instrumental measurements of textural properties.

Hardness recorded when sphere and cylinder were applied on fillets of equal thickness (2,0 cm) was higher than those from fillet samples of natural thickness at all locations on the fillet, except for location 6 for the sphere and location 7 for the cylinder (Fig. 5, 6, 7 and 8). Our results that hardness was different on different locations from head to tail confirmed results of Andersen (1995). However, they measured texture at only 3 locations on the fillets and they measured hardness or resistance against compression by a method based on puncture with a flat-ended cylinder.

Limited published information is available on texture measurements at different locations on fillets, but some indication of the importance of sampling technique has been published (Borresen, 1986; Botta, 1991). The main emphasis has been that the sample must be representative of the whole fillet. Azam et al. (1989) measured texture at 3 locations and used the means from the 3 measurements. Botta (1991) recommended use of measurements from 3 locations on cod fillets in order to maximize the correct assessment of fillets for sensory analyses. It may, be more feasible to apply texture measurements on locations which represent only part of the fillet.

Our results emphasize that the sampling technique is an important factor that can affect the final results of texture analysis. The difference in textural properties within one fillet can be higher in some instances than between fillets of different individuals. Mixing samples from different locations of a fillet may, therefore, be questionable. This can lead to difficulties in some cases in studying effects of processing or storage of fillets on texture. To study comparison of textural measurements and sensory evaluation of fillets after processing it is recommended that both fillets are processed first and the same locations on each fillet be compared in evaluations. Effects of storage or processing may be smaller than differences between locations on a fillet.

The cutting method showed more difference in textural properties (shear force) between locations on the fillet than the compression methods (hardness). This is an interesting alternative for studying the physical properties of the muscle, e.g., as related to the structure of the muscle. However, the compression method based on pressing 5 mm distance into the muscle is less destructive. The fillet samples do not have to be cut into equally thick parts and the fillet samples can be of natural thickness using duplicate samples at locations 2 to 4 (Fig. 1) for salmon of the equal sizes. When studying the tail part (locations 6 to 7), duplicates should not be used, but single samples at locations 6 or 7, because of inherent variation in textural properties at the tail part. The measurements of hardness would be more appropriate for following changes on the surface of the fillets rather than different textural properties. Dunajski (1979) recommended Kramer shear/compression cell to examine tenderness of fish muscle, however, large samples are needed. Therefore, other methods have been developed for measuring shear force such as by Chamberlain et al. (1993). However, the cutting method we applied is based on using smaller samples than for the Kramer shear/compression cell. Springiness was calculated from the TPA curve as the difference in puncture depth between the first and second compression. Springiness gave limited information and did not express significant results between different locations.

Sample size

Samples of salmon (50) were collected to investigate the effects of sample size on results and to give better understanding of the methodology. For evaluation and estimation of sample size that would be satisfactory for differentiation between population means, it was necessary to assume an approximate normal distribution of the population. We had to define the effect size (ES), i.e., the difference needed to
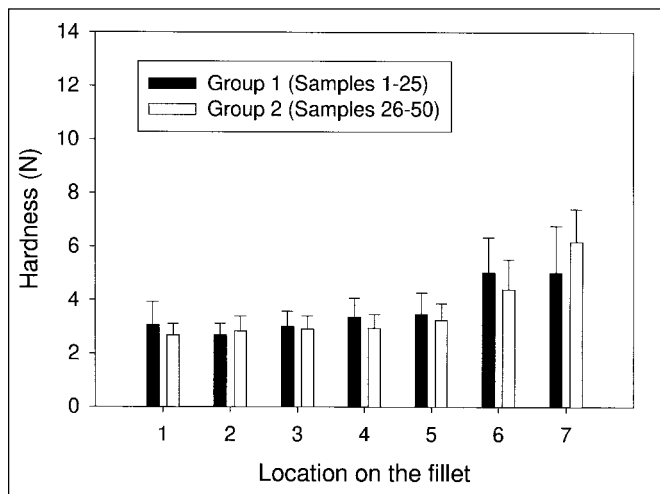

Fig. 8—Hardness of salmon fillets measured by using puncture with sphere at seven locations from the head to the tail. Samples were naturally (not equally) thick. Data are mean and standard deviation of 25 fish from the first sampling (1-25) and the second sampling (26-50). Location 6 and 7 are significantly different from locations 1, 2, 3, 4 and 5.
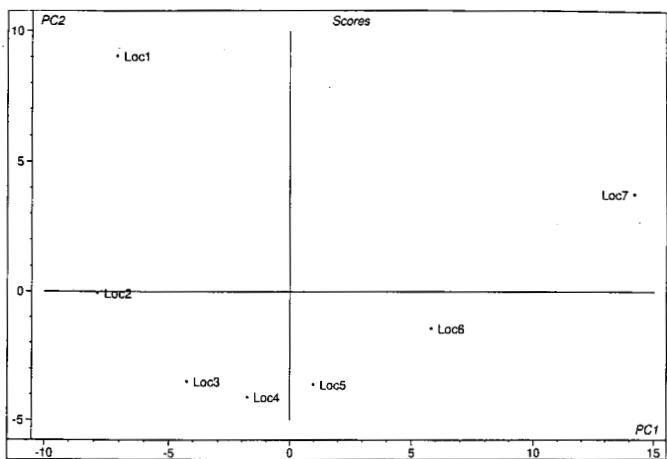

Fig. 9—PCA scores of the relation between the samples at seven locations from head to tail on salmon fillets within different texture methods (shear force by blade and hardness by cylinder and sphere for both equally thick samples and natural thickness).

Table 1—Thickness of salmon fillets from head (location 1) to the tail (location 7)[a]

Location (Fig. 1)	Right fillet cm	Left fillet cm
1	3.0 ± 0.3	2.8 ± 0.3
2	3.0 ± 0.3	3.0 ± 0.3
3	3.0 ± 0.3	2.9 ± 0.3
4	2.8 ± 0.3	2.7 ± 0.3
5	2.5 ± 0.3	2.6 ± 0.2
6	2.2 ± 0.3	2.3 ± 0.2
7	2.1 ± 0.2	2.2 ± 0.2

[a]Numbers are mean and standard deviation of 50 fish fillets.

Table 2—Regression of hardness/shear force as function of thickness of salmon fillets[a]

Sample	b[0]	b[1]	R²
Cylinder-samples equally thick	1402.3	−247.7	0.34
Cylinder-samples naturally thick	1708.0	−461.9	0.94
Sphere-samples equally thick	706.3	−62.5	0.20
Sphere-samples naturally thick	1084.1	−268.8	0.88
Blade-cut	14809.1	−4234.4	0.78

[a]Data are based on 50 fish fillets.

Table 3—Description of necessary sample size to discriminate between fillet samples with different alternatives of effect size for hardness and shear force measurements[a]

			Estimated sample size (no. of samples)			
			Equally thick samples		Naturally thick samples	
Want to separate hardness and shear force	ES %	Shear force cut	Cylinder	Sphere	Cylinder	Sphere
650g from 500g	26.1		12	9	14	9
650g from 600g	8.0		114	78	133	81
650g from 630g	3.1		742	501	865	530
400g from 300g	28.6		10	7	12	8
326g from 300g	8.3		105	71	122	75
4000g from 2000g	66.7	6				
3000g from 2000g	40.0	12				
3000g from 2500g	18.2	54				

[a]Data are based on 50 samples.

be able to separate the means, expressed as percent of the mean and to estimate the variation in the population, estimated by the sample coefficient of variation (CV). These calculations (Eq. 1) were based on the method by Nortvedt et al. (1996).

The CV and the ES had the greatest influence on the calculation. CV was generally not known for the population, but could be estimated from texture measurements performed. The sample size calculation was applied on the data from the shear force measurements, hardness from the sphere and the cylinder measurements on all fillets at location 3 (Table 3), because this would probably be most relevant in future measurements . The CV approached a lower threshold with increased sample size and the following results were obtained: CV=21.3% for the cylinder measurements (equally thick samples), CV=17.5% for the sphere measurements (equally thick samples), CV=23.0% for the cylinder measurements (naturally thick fillet samples), CV=18.0% for the sphere (naturally thick fillet samples) measurements and CV=33.3% for the shear force measurements.

The actual ES necessary for differentiation between averages would depend on the experimental design. If the aim was to discriminate between two fillets with mean texture hardness of 500 g and 650 g, respectively, the ES would be: [(650-500)/575]*100=26.1% (Table 3). A similar discrimination between 300g and 326g would give ES=[(326 - 300)/312]*100=8.3%. Using the sample size Eq (1), the required sample size, to be able to discriminate between samples with different alternatives of effect size for the two methods, are listed (Table 3). These values are based on the assumption that we wanted to be 80% sure that the estimated mean value was within a 95% confidence interval (P<0.05), i.e., the statistical power equals 0.8.

From this example, it was clear that the effect size had great influence on the necessary sample size. Any effort to reduce the CV in the preparation of the samples and in standardization of the measurements would be similarly effective. Although 50 samples were used in our study a larger sample than n=50 would be helpful in the estimation of a more reliable CV. Another way to reduce the sample size would be to reduce the demand of 80% statistical power or to increase the confidence interval around the mean (i.e. P<0.10), but this would not be as effective as improving CV or increasing the ES.

The number of samples applied in studies on textural measurements are often limited to 3 to 10 individual fish. That is one reason why limited significant differences often are achieved between sample groups. It is possible to use more than one sample from each fillet, but differences between individual fish studies on necessary sample size need to be done before evaluation of differences between sample groups.

Samples of different origin

Shear force was different between salmon fillets of different origins. Shear force of fillets from ocean-ranched salmon was higher than for farmed fish in land-based farms or fish from cages (Fig. 10). The ocean-ranched samples collected at end of the season in August were higher in shear force than the samples collected in June at the beginning of the season. Differences were observed at all locations from 1 to 7. The differences between samples is better represented using locations 5–7 than locations 2–4. This was as expected after performing all pair-wise multiple comparison procedures, where the number of differences in mean values among treatment groups were greater when examining the results from location 5–7 than 1-4, compared to those expected by chance (P<0.05). There was a statistically significant difference between 32 groups for location 7 but between 16 groups for location 1 although that was a higher standard deviation for location 7.

1 Sea-cages
2 Sea-cages
3 Landbased tanks
4 Sea-cages
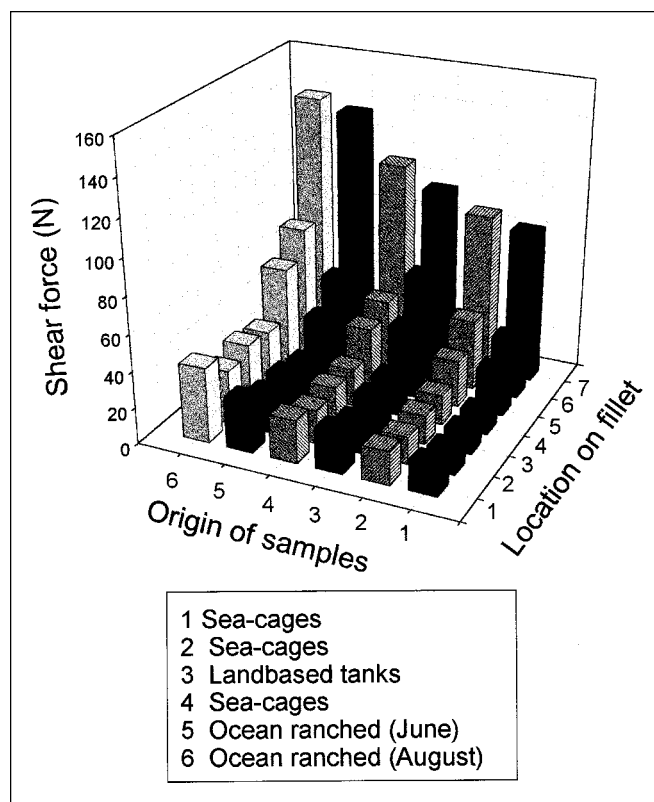5 Ocean ranched (June)
6 Ocean ranched (August)

Fig. 10—Comparison of shear force of salmon fillets of different origin, measured by blade on seven locations on the fillets using samples made equally thick. Data are mean of 15 fish samples.
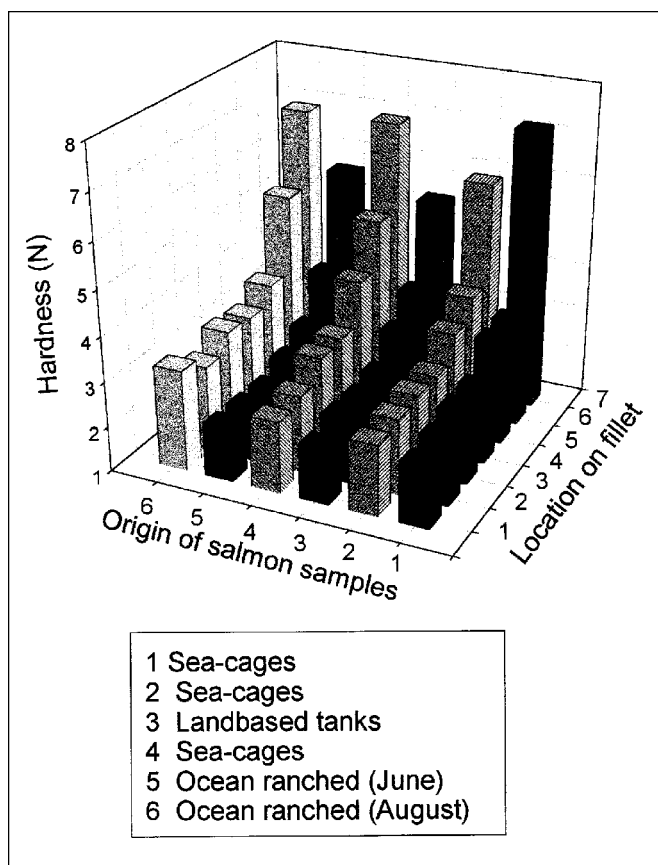
Fig. 11—Comparison of hardness of salmon fillets of different origin, measured by using puncture with sphere at seven location on the fillets. Data are mean of 15 fish samples.

Shear force of salmon fillets of different origin increased from head to tail which confirmed results from the experiment on sampling and sample size. No significant differences were observed between locations 1–4 in the samples of different origin. Hardness measured by the sphere showed that locations 1–4 were not different in any samples of different origin. Differences between samples of different origin were not as clear as shown with the cutting method with blade (Fig. 11). Ocean-ranched salmon (August) had higher hardness than all other samples at all locations from 1 to 6.

CONCLUSION

THE SHEAR FORCE METHOD BASED ON CUTTING WITH BLADE WAS more sensitive than the two compression methods and is recommended to use. However, the compression methods were less destructive than the shearing method. Hardness and shear force increased from head to tail. The location below the dorsal fin was the most reliable for practical applications.

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