

The influence of temperature on fish

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as measured by objective quality methods

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It is now clearly established that temperature is the most important factor influencing quality and shelf life of fish. An increase of only a few degrees in the vicinity of 0 deg. C considerably accelerates spoilage. Canadian workers found that cod (*Gadus morhua* L) spoils twice as quickly at 3 deg. C as at 0 deg. C.^{1, 2} Experiments by Sigurdsson³ on slightly salted herring (*Clupea harengus* L) gave similar results. Castell⁴ and Hansen⁵ concluded that lowering the temperature by a few tenths of a degree around 0 deg. C inhibited the growth of psychrophilic spoilage bacteria more than lowering the temperature by several degrees in the 10 deg. C range.

A comprehensive temperature survey carried out at all stages of fish handling and processing in Belgium showed that much more attention should be paid to this important problem. In many cases, the temperature of fish and shell fish was far too high and this greatly increased deterioration of the products involved.⁶

Within the programme for quality research by our laboratory, it was decided to study if temperature influences likely to occur in commercial practice could be followed by objective quality methods, and which determinations give the best response in this particular case.

Data on the evolution of spoilage at a continuously maintained storage temperature were presented by Hansen,⁵ Castell⁴ and Sigurdsson,⁷ who showed that, with several species of fish, total volatile bases, trimethylamine and volatile acids increased more rapidly at storage temperatures of 3 - 10 deg. C than at 0 deg. C.

Many less experiments are reported dealing with the exposure of fish to high temperatures during relatively short periods; the scarce data, moreover, refer to bacteriological determinations only. According to Reay and Shewan,⁸ storage life of cod is decreased by two to six days when the fish is exposed for 24 hours at 7 deg. C before being stowed in ice; bacterial load increased tenfold.

Riemann and Gramsnaes⁹ obtained similar results with fish allowed to stand for eight to 13 hours at 10 deg. C before being iced; shelf life was shortened by one to three days. Castell and MacCallum¹⁰ observed that keeping quality decreased by several days — especially in summer — if the fish remained unprotected on deck.

Preliminary experiments carried out by our Research Station have already been reported,¹¹ but they dealt only with cod and plaice exposed for 18 hours to different temperatures; total volatile bases, refractive index of the

eye fluid and electrical resistance of the fish flesh were determined.

In the work described here, the scope of the experiments was extended with regard to fish species and objective freshness tests.

Experiments

Objective quality methods: The following methods were used:

1. Refractive index (RI) of the eye fluid: according to Proctor *et al.*¹²

2. Electrical resistance of the fish flesh: by means of the Intelectron Fish-Tester V (Intelectron Electronics, Hamburg, Germany) developed by Hennings.¹³

3. Total volatile bases (TVB): by the method of Lücke and Geidel,¹⁴ but using Antonacopoulos' steam distillation unit;¹⁵ distillation time was 20 min.

4. Volatile reducing substances (VRS): by the method of Farber and Ferro.¹⁸

5. Trimethylamine (TMA): according to Dyer,¹⁷ but on 1 ml distillate of the TVB-method.

6. Volatile acids (VA): by the method of the AOAC,¹⁸ but using Antonacopoulos' distillation unit;¹⁵ 500 ml was distilled over.

7. Volatile ammonia: by the slightly modified micro-diffusion method of Vyncke and Merlevede;¹⁹ 20 g of fish is blended with 200 ml of distilled water for two min. and filtered. One ml of the filtrate is pipetted into a 50 ml microdiffusion flask; the polyethylene stopper is bored to fit a glass rod with ground extremity. One ml of saturated potassium carbonate is added; the glass rod is dipped into 1 N sulphuric acid and the excess shaken off. The stopper and rod are replaced on the bottle which is now rotated for 30 min. at 50 rpm. The rod is removed and dipped into 5 ml of diluted Nessler solution. Absorbance is read at 410 mμ.

Material: The following fish species were used:

— cod (*Gadus morhua* L): ca 2.5 kg. each, from the North Sea, period July - October, five days old;

— redfish (*Sebastes marinus* L): ca 1 kg. each, from Iceland, period September - November, five days old;

— herring (*Clupea harengus* L): ca 150 g. each, from the North Sea, period May - June (whole herring), three days old;

— dogfish (*Squalus acanthias* L): ca 1 kg. each, from the North Sea, period December - March, five days old; all

dogfish were beheaded and skinned before starting the experiments.

Great care was taken in choosing fish of approximately the same degree of freshness. When dealing with cod and redfish, fish giving Fish-Tester readings respectively of 35 - 45 and 25 - 35 were taken. Previous experiments indeed showed that the Intelectron Fish-Tester V is a laboratory instrument suited for that purpose;²⁰ 30 - 40 fish with a temperature between 0.5 and 1 deg. C were used for each experiment.

Procedure: All fish were divided into three lots. Lot A was spread on a wooden shelf and kept for 18 hours at 20 deg. C in a thermostatically controlled chamber. A second lot of fish (B) was kept in the same conditions at 15 deg. C, whereas a third lot (C) was stored directly in ice. After the 18 hours' period, all fish were packed in ice and stored at 1 deg. C.

Every two or three days, seven to ten fish of each lot were taken for analysis; 100 g. pieces were removed from the dorsal muscle of each fish and combined to one homogenous sample.

With cod and redfish, RI, Electrical resistance, TVB, TMA, VRS, and VA were determined. Only TVB, TMA, VRS and VA were taken into consideration with herring since RI cannot be measured and the electrical resistance of the fish was found to be unreliable.²⁰ With the skinned dogfish, RI and Fish-Tester cannot be used. VA and TVB are no more suitable methods, owing to the high urea content (1.5 - 2 per cent); upon distillation, ammonia and carbon dioxide are produced, which interfere with both determinations.

Besides VRS, it seemed worthwhile to test the applicability of the determination of volatile ammonia by the accelerated microdiffusion technique. In fact, preliminary experiments showed that this method could be used for

elasmobranch species since no hydrolysis occurs during the short diffusion time of 30 min.

In all cases, a simple sensory test was carried out. All experiments were repeated six times in the periods indicated above.

Results: In spite of unavoidable differences among the different fish samples, the general pattern of each experiment was very similar. For this reason, only the average values of the six experiments are mentioned in Tables 1 and 2.

Progress of deterioration appeared to be fairly similar when determined by the different objective quality methods under investigation.

With cod, all but one of the methods were able to distinguish between lots A, B and C.; RI however could only differentiate lots A and B, considered as a whole, and lot C. Results of the previous experiments with small and medium cod were thus confirmed and indicate that RI is not a sensitive method.

Fish-Tester readings (Q-values) of the three lots decreased markedly the first two days. After eight days, moreover, lots A and B reached the O-limit. This would imply that electrical resistance of the fish flesh is strongly influenced by a raise of temperature and would confirm Hennings' observations,¹³ that measurements on fish allowed to warm up to 10 deg. C, are not reliable.

It should be emphasized in this connection that Fish-Tester readings given by different apparatus or given by the same apparatus at wide spaces of time can only be compared if the calibration of the units is carefully checked.

Three years' experience with the new device indicates that marked variations can occur between fish of the same organoleptic quality due only to an altered calibration. Very low Q-values should be particularly suspect.

Table I. — Influence of three temperatures (A, B, C) on the spoilage of cod (*Gadus morhua* L), redfish (*Sebastes marinus* L), and herring *Clupea harengus* L) as measured by six objective quality methods.

Determination	Lot	Cod				Redfish				Herring			
		0d	2d	5d	8d	0d	2d	5d	8d	0d	2d	4d	7d
RI (*)	A	66	88	94	108	64	63	72	80	—	—	—	—
	B	66	89	99	97	64	68	73	77	—	—	—	—
	C	66	76	81	91	64	64	68	70	—	—	—	—
Fish-Tester (Q)	A	38	10	5	0	30	12	8	0	—	—	—	—
	B	38	12	6	0	30	15	10	5	—	—	—	—
	C	38	22	14	7	30	32	17	11	—	—	—	—
RVB (mg N %)	A	27.0	43.3	61.0	84.4	22.4	27.3	38.1	55.5	26.1	39.6	48.2	70.0
	B	27.0	34.5	47.7	75.3	22.4	25.4	33.2	46.5	26.1	35.6	42.8	66.2
	C	27.0	29.1	34.3	53.2	22.4	23.1	30.0	34.0	26.1	26.8	28.4	40.8
TMA (mg N %)	A	3.7	14.7	27.1	39.3	2.7	5.0	14.6	23.3	1.3	7.0	15.3	24.0
	B	3.7	9.5	19.3	32.0	2.7	3.9	9.4	16.5	1.3	6.6	12.6	22.6
	C	3.7	4.4	8.3	16.4	2.7	3.0	7.1	10.6	1.3	1.5	2.9	11.3
VA (ml 0.01 N NaOH per 100g)	A	29.7	64.8	108.0	160.8	25.3	37.2	56.4	106.8	13.1	34.2	49.1	67.5
	B	29.7	46.8	76.8	132.0	25.3	33.6	54.0	79.2	13.1	30.8	44.5	65.2
	C	29.7	33.6	38.0	84.0	25.3	26.4	43.2	57.6	13.1	15.2	22.8	39.1
VRS (µeq 5 ml juice)	A	10.1	25.6	38.7	40.8	7.3	13.8	19.1	34.3	6.0	17.0	26.6	36.2
	B	10.1	20.2	36.2	37.9	7.3	11.4	15.8	31.5	6.0	14.2	22.9	33.7
	C	10.1	11.6	25.8	31.4	7.3	8.3	12.9	20.0	6.0	6.2	8.7	19.7

(*) Expressed as (RI - 1.3300) 10⁴; e.g. 1.3366 giving 66.

Table 2. — Influence of three temperatures (A, B, C) on the spoilage of dogfish (*Squalus acanthias* L) as measured by two objective quality methods.

Determination	Lot	Fig:d	2d	5d	8d
NH ₃ (mg N %)	A	25.8	70.2	87.5	139.6
	B	25.8	36.9	69.9	104.8
	C	25.8	29.0	48.2	50.4
VRS (µeq/5ml. juice)	A	8.6	25.9	41.6	44.3
	B	8.6	20.6	24.0	36.2
	C	8.6	10.9	10.6	20.9

With redfish, RI appeared to be a poor method; individual data (not quoted) moreover showed large fluctuations especially in lot A. These findings confirm previous results:^{11, 12} RI is not suitable for redfish. In contrast with cod, Fish-Tester values decreased more slowly and only lot A reached the lower limit ($Q=0$) after eight days. This would point out that Q -values of redfish are less influenced by a temporary raise in temperature. The other methods, (TVB, TMA, VA and VRS) were able clearly to differentiate the three lots.

This also occurred with herring, but average spoilage values of lots A and B increased more rapidly, indicating a greater sensitiveness to temperature influences.

With dogfish, VRS and volatile ammonia could draw a clear distinction between the three lots.

It should be noticed that the accelerated microdiffusion method determines free ammonia produced during degradation of proteins, free peptides and aminoacids as well as free ammonia from urea hydrolysis. Since this latter is due almost entirely to bacterial urease²¹ it can be considered as a spoilage product. This is also strongly suggested by the organoleptic judgment which was in good agreement with the determination of volatile ammonia.

The VRS-method on the other hand is not influenced by free ammonia, but determines other volatile components (volatile acids, aldehydes, ketones, alcohols, amines, etc.).¹⁶ Hence, both methods appeared to be of real value for the objective quality determination of elasmobranch fish, especially if it is borne in mind that very few methods are suitable for those fish.

The reported data (Tables 1 and 2) also give some indication on the relative sensitivity of the four fish species against temperature influences. When examining the different spoilage patterns of the teleost fish, it can be observed that redfish is the least and herring the most sensitive fish, while cod takes an intermediate position.

All spoilage values of herring lots A and B increased sharply after only two days of storage. With redfish on the other hand, objective quality determinations on lots A and B gave less varied results. These findings are in good agreement with the known fairly high resistance of redfish to spoilage.

Dogfish, the only elasmobranch fish tested, is also very sensitive to temperature influences: after only two days a marked difference between the three lots occurred.

In these experiments, stress was laid on the comparative testing of the objective quality methods mentioned. The results, however, allow us to draw some conclusions with regard to temperature itself.

The temperature influences described here often occur

in commercial practice. After unloading, the fish remains without cold protection for about ten hours in the auction hall.⁶ Afterwards, it often remains unprotected for hours in the premises of wholesale and retail traders. The detrimental influence of temperature on quality and shelf life of fish appears clearly and is especially characteristic of fish which is completely exposed to the influence of surrounding warm air; this occurs, for instance, when fish is exposed on the floor in the auction hall, when it is spread out on tables by the wholesale trade, or when it is displayed in unrefrigerated windows of fish shops.

Finally, these experiments confirm previous results obtained with temperature and packing experiments.²²

Summary: The influence of three temperatures (20 deg., 15 deg. and 0 deg. C) during 18 hours on the spoilage of cod (*Gadus morhua* L), redfish (*Sebastes marinus* L), herring (*Clupea harengus* L) and dogfish (*Squalus acanthias* L) was tested by means of the following objective quality methods: refractive index (RI) of the eye fluid, electrical resistance of fish flesh, total volatile bases, trimethylamine, volatile reducing substances (VRS) volatile acids and volatile ammonia.

All methods — RI excepted — were able clearly to differentiate the above mentioned temperature influences. With dogfish, VRS and volatile ammonia determinations proved to be of real value for quality assessment.

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