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The blood content in fillets of fish after mechanical filleting.

The effect of delayed filleting for one or more hours without previous gutting.

A comparison between the effect of gutting and of direct eviscerating quite fresh saithe.

by

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The rapid development of the fish-catching technology on board trawlers has led to that the gutting process previously used, seems too time-consuming for factory trawlers.

Investigations have been necessary to adapt the complete removal of blood to the mechanical eviscerating procedure.

Methods

For preparation of mechanical eviscerated fillets small saithe (Gadus virens) were used. They were killed and gutted immediately before filleting on a Baader machine no 181 and a Baader skinning machine no 47.

For investigation of the effect of storage at 0°C before the fish was eviscerated and the blood removed, living small saithe were used.

For comparison of gutted to directly eviscerated newly caught fish, living small saithe (½ kg) were gutted respectively eviscerated in a narrow vessels, where the blood haemolysed into 500 ml water without any coagulation.

The amount of exuded blood was measured as alkaline haematin (Lawrie, 1950). To 5 ml blood-water (exuded blood into 500 ml water) was added 5 ml 2 N HCl. After 40 minutes 10 ml 2 N NaOH was added. The extinction at 605 mμ was measured after 30 minutes.

The amount of blood was calculated according to the Haden hemoglobin standard. This contains 0,522 mg Fe in 100 ml, which represents the normal heamin content of blood diluted 1:100 with 0,1 N NaOH.

For the calibration curve haemin prepared from ox-blood after the method of Delory (1943) was used and purified according to the method of Fisher (1941). The curve was checked with different trade products.

The hemoglobin content in blood of saithe was calculated as being the same as in man (15,6 g/100 ml blood) (Prosser, 1952).

The very small amount of blood in the fillets was measured by converting the hemoglobin into protoporphyrin with hydrazindihydrochloride (Chu and Chu, 1953). Blood had by this method to be diluted 1:10.000.

1 ml diluted blood + 3 ml reagent A were heated in boiling water for 3 minutes. Reagent A: 1,0 g hydrazine-dihydrochloride was diluted in 50 ml conc. acetic acid and 50 ml 0,1 N HCl.

The fluorescence was measured with Perkin-Elmer Fluorescence Spectrophotometer MPF-2A at 600 mμ. For extraction of blood from the fillets, these were washed and grinded. For extraction of blood was tried a) 50 g minced muscle homogenized with 50 ml 0,1 % K-oxalate, b) 50 g minced muscle homogenized with 50 ml water.

Table 1 shows that both methods gave the same value for blood in cod.

Standard-solutions for fluorometric measurements

Fish blood was diluted to the range 0,025 - 1 mg/ml. Suitable concentrations for measurements lay between 0 and 0,5 mg blood/ml (Table 2).

In the case the solutions of blood was too concentrated, the emission was hindered. As the spectrophotometer appeared to be unstable, standards were always measured together with the samples.

By using the standard solution of Haden diluted 1:100, that means 0,156 g hemoglobin in 100 ml 10 % acetic acid the measurement of fluorescence gave nearly the same curve (Table 3). The corresponding standard-solution of Haldane contained 13,8 and of Sahli 17,3 g hemoglobin (Clegg and King, 1942).

A mixture of the standard solution and the sample gave also corresponding results.

Results

Investigations of the content of blood in fillets of fish after mechanical filleting

The content of blood in fillets of fresh saithe is shown in Table 4 and 5. The 16 fishes in Table 4 are caught at the same time and are nearly of the same size. The fillets are somewhat above 200 g. The average blood content is 0,66 ml/kg fillet and the middle deviation $\pm$ 0,01 ml.

Table 5 represents 24 fishes caught at the same time. These fishes were smaller, and the fillets were generally under 200 g. The average blood content is 0,45 ml blood/kg fillets and the middle deviation $\pm$ 0,03 ml.

The effect of delayed filleting for one or more hours without previous gutting

A series of measurements of saithe from the same catch were carried out. The fish was filleted after three hours (Table 6), respectively after four days at 0° without being gutted (Table 7).

After three hours the middle blood content was 0,83 ml per kg fillet with a middle deviation of $\pm$ 0,02. After four days the content was 0,95 ml $\pm$ 0,06.

In another series the amount of blood in the fillet after 1 hour at 0° was 0,40 ml/kg (Table 8), after two hours 0,44, after five hours 0,52 and after 24 hours 0,53 ml/kg.

The corresponding total amount of blood in the fish stored 24 hours at 0°C varied between 26 and 41 ml per kg fish with an average of 30,9 ml.

A comparison between the effect of gutting and of direct eviscerating quite fresh fish

The blood content of fillets of saithe of the same shoal was measured. In this experiment (Table 13) with 12 fishes the middle value was the same in the case of gutting and of direct eviscerating.

Also the amount of excuded blood from the fish (Table 14) was nearly the same, 10,9 ml/kg fish.

The amount of excuded blood was here also estimated with alkaline hematin method. The values are only a little lower (Table 15).

In another experiment with three fishes gutted and three eviscerated, the remaining blood in the fillets was a little higher in the gutted fishes (Table 16), but on the other side also the excuded blood was higher in the gutted fishes (Table 17).

Also in this case there was correspondence between the two methods of estimation of blood.

Discussion

The amount of blood in fishes is influenced by several factors.

The blood volume in "la plie de la Baltique" is found to be 27,2 ml/kg, Buddenbrock, Henschel and Friedrich (1934), in Carpe 28,5, Fontaine et al. (1945).

In more active fishes the blood volume is according to M. Fontaine (1958) still higher.

The number of red blood cells (erythrocytes) is depending on size and sex-cyclus. The temperature of the water seems to be of great importance. K. Hashimoto (1963), Scholander and Dam (1957).

Our measurements of the total amount of the blood-volume lay between 26 and 41 ml/kg with a middle value of 30,9 ml, which is only a little higher than in carp. As standard for the hemoglobin content of saithe has been used the "Haden" standard (15,6 g hemoglobin/100 ml), which is close to the human hemoglobin content.

The use of this standard is supported by the measurements of Table 2 and 3. These show that a standard curve with blood of saithe (Table 3) corresponded well with the curve with "Haden" standard (Table 2).

Prosser (1952) showed great variations in the hemoglobin content of fishes between the different species, above and under our value.

Widemeyer and Chatterton (1971) calculated with 6,5-9,9 g hemoglobin/100 ml in salmon.

Another factor which has not been sufficiently investigated is the variation in blood clotting-time (Macnab and Ronald 1964), which may influence the results.

To obtain the best possible basis for comparison in our experiment, fishes of the same age, the same season and the same shoal have been used.

Mechanical filleting

There is a clear difference in blood content between the two catches, Table 4 and 5. One of the reasons may be a difference in size of the fillets. The fillets (Table 4) with an average blood content of 0,66 ml/kg were somewhat above 200 g. The fillets in Table 5 however, have an average blood content of 0,45 ml/kg. These fillets were generally under 200 g. The explanation why the greatest fillets have the highest blood content may be: a) the thickness of the fillets hinder exudation of the blood, b) the greater fishes contain more hemoglobin.

Delayed filleting

Table 6 and 7 show the effect of delayed filleting at $0^{\circ}C$ for 3 hours and for 4 days on fish of the same size and the same shoal ($0,83 \pm 0,02$ ml blood/kg fillet and $0,95 \pm 0,06$ ml blood respectively). Table 8, 9, 10, 11 represent another investigation with fish of the same size but another shoal.

The blood content in the fillets increased slowly from 0,40 ml/kg after 1 hour delay to 0,44 ml after 2 hours, 0,52 ml after 5 hours and 0,53 after 24 hours.

The great difference in the blood content of the fillets of these two investigations is remarkable, further the small difference between 1 and 24 hours (Table 8 and 11).

Comparison between gutting and evisceration

In accordance with our earlier investigations (Jebsen, 1972) the effect of removing the blood by direct eviscerating and by gutting gave nearly the same result both in amount of blood remaining in the fillets (Table 13) and the blood removed from the fish (Table 14).

In another experiment with 6 fishes, the gutted fishes showed somewhat higher content of remaining blood (Table 16) simultaneously the amount of exuded blood from the fish was higher than in the directly eviscerated fishes (Table 17). This may be explained that in this case the three gutted fishes possessed a total higher blood-volume than the corresponding eviscerated fishes. As shown in Table 12 there may occur great difference in total blood volume.

Summary

The investigation was carried out with fluorometric porphyrin-determinations and alkaline haematin-determinations which gave corresponding results.

By mechanical filleting the blood content in the fillets showed small variations between fish of the same catch, but between different catches and seasons the values could vary.

By storing the fish ungutted for three or more hours at $0^{\circ}C$ the blood content increased, though not in the expected degree. It was lower than the variations between fresh fillets from one catch to another.

A comparison between gutting and direct eviscerating without previous gutting showed that in both cases the blood content which remained in the fillets was the same, and the blood exuded from the fish was the same.

Table 1 Extraction of blood from cod and saithe with K-oxalateor water

Solution of extraction	Sample	Relative emission 600mμ at degree of sensitivity 4	5	6	ml blood per ml sample	ml blood per kg fish
K-oxalate	Cod		35,5		0,090	0,18
H_2O	Cod		34,2		0,090	0,18
Standard 1		69,5			1,0	
" 2			39,9		0,1	
" 3				28,4	0,05	
" 4				13,0	0,025	
K-oxalate	Saithe		70,8		0,32	
H_2O	Saithe	32,2	96,6		0,53	0,86
Standard 1		60,0			1,0	
" 2			32,0		0,1	
" 3				20,1	0,05	
" 4				7,9	0,025	

Table 2 Fluorometric standard solution from saithe-blood

Standard solution	ml blodd per ml solution	Relative emission by 600 mμ at degree of sensitivity 4	5
S 1	0,1	16	48
S 2	0,2	20	65
S 3	0,3	24	82
S 4	0,4	41	
1 ml blood-sample	0,345	30	
0,5 ml blood sample + 0,5 ml standard S 1)		24	83

Table 3 Fluorometric "Haden" standard solution 0,156 g hemoglobin in 100 ml acetic acid solution 10 %

Standard solution	Haden standard ml blood per ml solution	Relative emission by 600 mμ at degree of sensitivity	
		4	5
S 1	0,1	13	43
S 2	0,2	23	78
S 3	0,3	29	(97)
S 4	0,4	30	

Table 4 Content of blood in fillets over 200 g of saithe. The fillets were from completely fresh fish, gutted short time before mechanical filleting (catch no 2)

Sample no	ml blood per kg fillets
1	0,69
2	0,73
3	0,73
4	0,73
5	0,64
6	0,70
7	0,68
8	0,63
9	0,58
10	0,70
11	0,58
12	0,61
13	0,60
14	0,66
15	0,63
16	0,64
Mean	0,66 $\pm$ 0,01

Table 5 Content of blood in fillets under 200 g of saithe. The fillets were from completely fresh fish, gutted short time before mechanical filleting (catch no 3)

Sample no	ml blood per kg fillets
1	0,34
2	0,45
3	0,44
4	0,50
5	0,44
6	0,67
7	0,45
8	0,40
9	0,53
10	0,41
11	0,40
12	0,57
13	0,72
14	0,54
15	0,31
16	0,39
17	0,61
18	0,20
19	0,53
20	0,29
21	0,37
22	0,31
23	0,37
24	0,44
Mean	0,45 $\pm$ 0,03

Table 6 Blood content in fillets after 3 hours unbleeded storage at 0°C before filleting (catch no 4)

Sample no	ml blood per kg fillets
1	0,85
2	0,78
3	0,82
4	0,86
5	0,88
6	0,78
7	0,66
8	0,82
9	0,88
10	0,88
11	0,81
12	0,77
13	0,93
14	0,92
Mean	0,83 $\pm$ 0,02

Table 7 Blood content in fillets after 4 days unbleeded storage at 0°C before filleting (catch no 4)

Sample no	ml blood per kg fillets
1	1,21
2	1,04
3	0,98
4	1,21
5	1,16
6	0,86
7	0,76
8	1,21
9	1,04
10	0,76
11	0,54
12	0,60
13	0,90
14	1,04
Mean	0,95 $\pm$ 0,06

Table 8 Blood content in fillets after 1 hour unbleeded storage at 0°C before filleting (catch no 5)

Sample no	ml blood per kg fillets
1	0,45
2	0,44
3	0,33
4	0,36
Mean	0,40 ± 0,03

Table 9 Blood content in fillets after 2 hours unbleeded storage at 0°C before filleting (catch no 5)

Sample no	ml blood per kg fillets
5	0,49
6	0,51
7	0,37
8	0,37
Mean	0,44 ± 0,04

Table 10 Blood content in fillets after 5 hours unbleeded storage at 0°C before filleting (catch no 5)

Sample no	ml blood per kg fillets
9	0,44
:0	0,61
11	0,59
12	0,46
Mean	0,52 ± 0,04

Table 11 Blood content in fillets after 24 hours unbleeded storage at 0°C before filleting (catch no 5)

Sample no	ml blood per kg fillets
13	0,49
14	0,56
15	0,56
16	0,40
Mean	0,53 ± 0,04

Table 12 Blood content in whole saithe. Stored unbleeded for 24 hours at 0°C. (catch no 5)

Sample no	Weight of the fish	ml blood per kg fish
17	600 g	41,0
18	640 g	30,5
19	580 g	26,0
20	510 g	26,0
Mean		30,9 ± 3,5

Table 13 Blood content in fillets of fresh saithe, gutted (A) or eviscerated directly (B) (catch no 6)

Sample no	ml blood per kg fish
A 1	0,48
A 2	0,48
A 3	0,84
A 4	0,38
A 5	0,42
A 6	0,50
Mean	0,51 ± 0,07
B 1	0,49
B 2	0,47
B 3	0,30
B 4	0,52
B 5	0,73
Mean	0,50 ± 0,07

Table 14 Exuded blood by gutting (C, A) or by direct eviscerating (C, B) (catch no 6, the same fishes as in table 13).
Fluorometric determination

Sample no	ml blood per kg fish
C, A 1	8,6
C, A 2	9,6
C, A 3	10,5
C, A 4	7,1
C, A 5	14,5
C, A 6	11,3
Mean	10,3 ± 1,0
C, B 1	11,5
C, B 2	13,5
C, B 3	9,2
C, B 4	9,6
C, B 5	7,6
C, B 6	13,8
Mean	10,9 ± 1,0

Table 15 Exuded blood by gutting (C, A) or by direct eviscerating (C, B) (catch no 6, the same fishes as in table 14)
Colorimetric determination

Sample no	Weight of the fish	ml blood per kg fish
C, A 1	551 g	7,2
C, A 2	627 g	7,4
C, A 3	591 g	8,9
C, A 4	562 g	5,2
C, A 5	516 g	9,3
C, A 6	553 g	11,7
Mean	560 g	8,3 ± 0,90
C, B 1	661 g	8,7
C, B 2	689 g	10,0
C, B 3	597 g	8,4
C, B 4	612 g	8,0
C, B 5	641 g	5,9
C, B 6	614 g	11,4
Mean	635 g	8,7 ± 0,8

Table 16 Blood content in fillets of fresh saithe, gutted (A) or eviscerated directly (B) (catch no 7)

Sample no	Weight of the fish	ml blood per kg fish
A 7	669 g	0,52
A 8	550 g	0,47
A 9	550 g	0,52
B 7	542 g	0,38
B 8	529 g	0,40
B 9	563 g	0,48

Table 17 Exuded blood by gutting (C, A) or by direct eviscerating (C, B) (catch no 7, the same fishes as in Table 16)

Sample no	Fluorometric determination ml blood per kg fish	Colorimetric determination ml blood per kg fish
C, A 7	12,9	11,1
C, A 8	19,3	10,9
C, A 9	16,9	10,0
C, B 7	8,1	7,9
C, B 8	10,4	8,6
C, B 9	7,8	8,2

Litterature

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